Message from the Director

We are pleased to announce that the Accreditation Committee of the American Association of Veterinary Laboratory Diagnosticians (AAVLD) informed the NCVDLS in early November that it had voted to grant full accreditation status to our laboratories through December 31, 2018.

The purpose of AAVLD accreditation is to accredit public veterinary diagnostic laboratories in North America relative to technical and operational competence. The AAVLD quality assurance standards for its member laboratories incorporates by reference the World Organization for Animal Health (Office International des Epizooties) (OIE) document "OIE Quality Standard and Guidelines for Veterinary Laboratories: Infectious Diseases," the international standard for laboratory quality. An accredited laboratory is one that is capable of providing a full range of diagnostic services to include bacteriology, mycology, virology, parasitology, serology, toxicology, necropsy, and histopathology testing services.

Two members of the accreditation committee and one subject matter expert auditor conducted the site visit and spent 4 full days auditing our facilities to review quality-assurance and quality system standard operating procedures, adequacy of equipment, employee training records, financial status of the laboratory and information technology policies. The audit team also met with members of the external laboratory advisory committee, the State Veterinarian and the Deputy Commissioner of Agriculture.

A quality assurance program in a veterinary diagnostic laboratory is essential to provide clients with a sense of confidence in the laboratory's test results and services. Clients who receive timely test results and are confident in their accuracy and reliability are empowered to make valid medical decisions involving the care of their patients.
A new option for transmitting laboratory reports is now available to our clients. We now have the ability to e-mail reports as PDF attachments. In the past, a link to our website was e-mailed to notify clients that a report was ready for them to view. Since e-mailing reports is an unsecure method of distribution and because the laboratory has a responsibility to protect client confidentiality, we are requiring clients to sign a waiver of acknowledgment prior to changing their report distribution status to e-mail. Please contact Ms. Tamara Seago at the Rollins Laboratory (919-733-3986) if you wish to make this change.

### Feature Article

**Substantial Winter Livestock Losses in North Carolina**

Drs. Jennifer Haugland, Mahogany Wade-Caesar, and Alison Tucker – Rollins Laboratory

The unusually wet and cold winter is having a significantly negative impact on livestock throughout North Carolina. The North Carolina Veterinary Diagnostic Laboratory System laboratories are seeing an increase in cases of starvation and parasitism in livestock species. Necropsy submissions since December 1, 2013, are up nearly 13% compared to the same period last year and starvation diagnoses have nearly doubled, from 11% last year to 19% of submissions this year. Cases with gastrointestinal parasitism have also contributed to more deaths this winter than previous years.

Starvation is unfortunately a common diagnosis in the livestock and sometimes companion animals submitted to the NCVDLS. A diagnosis of starvation means the daily consumption of energy and protein was inadequate to maintain body condition. Factors such as parasitism, poor teeth, and chronic disease may contribute to poor body condition but starvation is the lack of adequate nutrition. The cold and wet weather greatly increases the energy demands of all species. Animals have a critical temperature point, which is the minimum temperature that requires no additional energy to maintain body condition. When the ambient temperature (including the wind chill) falls below the critical temperature, animals need to consume additional energy every day to keep from losing weight. For cattle with a dry winter coat, the critical temperature is 32 F degrees. So for every degree below 32 F, 1% more energy (TDN) in the daily ration is required to prevent weight loss and that is in addition to gestational or lactational needs. However, during this winter cattle are often wet and when they have a wet winter coat the critical temperature increases to 60F! So on these wet sloppy 35 F winter days; cattle need an additional 25% of energy every day. If there is no protection from the wind, the ambient temperature can be a lot lower. The owners are feeding their animals but hay, minerals, and protein tubs are not meeting the metabolic needs. So metabolism becomes catabolic which mobilizes fat and reduces muscle mass leading to severe weight loss and death.
From the fall of 2013 to current, an increased number of gastrointestinal parasite related deaths in cattle submitted to the North Carolina Veterinary Diagnostic Laboratory has been observed in comparison to previous years. The majority of these animals are young; however older calves and mature adults have been affected with significant parasitism. An increased presence of *Oesophagostomum* sp., which causes many pea-sized black nodules (granulomas) along the intestinal mucosa, has been observed. Also the barberpole striped nematode has been observed along the abomasal mucosa of several calves. These worms were confirmed to be *Haemonchus* sp. by Dr. Bruce Hammerberg, parasitologist at NCSU-CVM. In some cases, the numbers of *Haemonchus* worms were enough to cause clinical anemia. The diagnosticians at Rollins Laboratory have not found *Haemonchus* in calves in previous years, although *H. contortus* is commonly present in small ruminants. Further speciation of these worms would tell us if there is an increase presence of *Haemonchus placei*, which commonly infects cattle, or are we seeing the results of comingling cattle and small ruminants.

NCVDLS veterinarians are recommending that owners contact their veterinarian or county extension agent to assess the nutritional needs of their livestock based on their body condition, their stage of production, and their weather exposure and to evaluate the need for mid-winter deworming. In a herd that is suffering from starvation, not all animals may look emaciated. The marginal ones, such as the young, old and timid, will be most severely affected. Starvation is a real diagnosis that warrants immediate action and parasitism may be more significant this winter. A more in-depth discussion is provided in Animal Science Fats publication [ANS 03-001 B, A Ten-point Plan for Winter Feeding Beef Cows](https://www.ncsu.edu/extension/bovine/index.html), by Dr. Matthew H. Poore, and we encourage veterinarians and producers to view the [NC Cooperative Extension website](https://www.ncsu.edu/extension/) for a host of articles on livestock management and nutrition.

### Short Cuts

**COMPANION ANIMAL**

**Canine**

A kennel of hunting dogs lost 2 puppies that were about 6 months old. The second puppy was diagnosed with acute renal failure, was euthanized and then submitted for necropsy. At necropsy the kidneys were enlarged and each had a heavily vascularized capsule. There were multiple to coalescing white and firm foci throughout the cortex and medulla of each kidney. Histopathology revealed marked neutrophilic and lymphohistiocytic tubulointerstitial nephritis, which is very suggestive of *leptospirosis*. Serology revealed an elevated titer (>1:6400) of only one *Leptospira* serovar, grippotyphosa. Immunohistochemistry of kidney also confirmed the presence of these organisms. This puppy and several others had been housed in an outdoor pen their whole lives and had never been out of the pen. *Leptospira* is transmitted in the urine of infected animals, including rodents which may have access to animal feed and water. *Leptospira* may also cause disease in humans when they are exposed to the urine of infected animals.

Dr. Jennifer Haugland
Cattle

An approximate 727 kg, four year-old, Angus bull is presented for necropsy after a five day history of increased respiratory rate, and then acute death. The bull was treated with anti-inflammatory medications and multiple antibiotics but did not improve. On necropsy, the bull was in ideal body condition, was mildly dehydrated, and had moderate tissue autolysis present. There was significant emphysema present in the subcutaneous tissues, and around the liver and kidneys. The cranial lung lobes were dark red, and were wet and rubbery on palpation. The periphery of these lobes had significant emphysema present. The caudal lung lobes were light pink to tan in color, were soft and spongy on palpation, and had significant emphysema present throughout the entirety of the lobe. The heart, liver, kidneys, and digestive tract were all unremarkable grossly. Histopathology of the affected lung tissue showed marked acute interstitial pneumonia with type I pneumocyte necrosis, hyaline membranes, type II pneumocyte hyperplasia and edema. **Atypical Interstitial Pneumonia** is typically seen in adult cattle that are moved from a dry to very lush pasture. Lush grasses can be high in the amino acid L-tryptophan, which is converted by ruminal microorganisms to indole acetic acid and then to 3-methyl-indole (3MI), which is toxic. The disease is also known as “Fog Fever” because the most lush grass is in the valleys with the fog. The disease can also occur with the ingestion of Perilla mint, moldy hay / silage, or sweet potatoes. The mold causes a hypersensitivity reaction within the lungs and results in damage to the pneumocytes. The disease can been seen in calves that are infected with Bovine Respiratory Syncytial virus. In this particular case, the bull was fed hay that was known to have mold in it.

Dr Brad Barlow

Caprine

Three 2 to 3-year-old goats from separate farms were submitted for necropsy due to neurologic signs characterized by ataxia, blindness, circling, cranial nerve deficits, paralysis, inability to swallow, and/or recumbency. Gross examination of the brain and spinal cord was unremarkable. On meningeal and cut section, the cerebrum of each animal did not fluorescence under ultraviolet light. The brains were negative for rabies virus via fluorescent antibody testing. Histologic examination of the brainstems and spinal cords revealed lymphohistiocytic perivascular meningoencephalomyelitis with neuropil necrosis and microabscesses, consistent with **listeriosis**. Listeriosis, caused by *Listeria monocytogenes*, can affect ruminants, other animal species, and humans (zoonotic). Animals of any age and sex may be affected; however animals less than three years of age are more commonly prone to clinical disease than older animals. The encephalitic form is mostly seen in adult animals and the septicemic form usually occurs in neonates. Intrauterine infection of the fetus can result in abortion in cattle and sheep. *L. monocytogenes* is transmitted from animal to animal through fecal oral routes, usually via manure contamination of the pasture or silage with the microorganism.

Dr. Mahogany Caesar
Caprine

Hypothermia, drooling, confusion, recumbency, inappetence, stranguria and anuria were observed in a 2-year-old male castrated Nubian/Nigerian dwarf goat. On necropsy examination, urethral obstruction with a ruptured urinary bladder was diagnosed. The abdominal cavity was fluctuant and filled with 3.6 liters hemorrhagic urine admixed with large blood clots. The apex of the urinary bladder was ruptured and large blood clots were adhered to the rupture site. The tip of the penis was necrotic and the associated urethra was completely obstructed by sandy material admixed with blood and mucous. This animal ultimately died from uremia. Analysis of the sandy material by the Minnesota Urolith Center revealed a composition of 80% magnesium ammonium phosphate (struvite) and 20% magnesium calcium phosphate apatite form. **Urolithiasis** in ruminants occurs as a result of nutritional disease. Dietary mineral content is critical, as there is a relationship between the type of diet fed and the type of urolith formed. For instance, high-grain diets with low calcium:phosphorus ratio increases the risk of developing struvite uroliths. The best way to prevent urolithiasis is to maintain appropriate dietary mineral levels (ie. magnesium, phosphorus, and calcium) for the age and development status of the animal. Note that the Minnesota Urolith Center performs urolith analysis free of charge; more information about the testing may be found on their website.

Dr. Mahogany Caesar

Camelid

A 1.5 year old male miniature llama from Cabarrus County examined at Griffin Animal Disease Laboratory had been lethargic for several days and died despite veterinary administered treatments. This llama had been purchased and transported to the Carolinas at about 3 months of age. Body weight was about 90 kilograms. Approximately 1.5 milliliters of red tinged fluid was in the abdominal cavity. The liver was swollen, tan to light reddish brown on the cut surface with numerous liver flukes and associated migratory tracts on the cut surface. Histopathology findings of the liver included severe neutrophilic and lymphoplasmacytic hepatitis, chronic with abscesses and marked fibrosis. The fluke species was identified as **Fascioloides magna**. **Fascioloides magna** is the Giant American Liver Fluke or Deer Fluke. The normal definitive host for **Fascioloides magna** is commonly the Whitetail Deer as well as other cervid species. The fluke eggs are shed in the feces of deer and in a moist environment will hatch and enter into a snail host. After further development in the snail, another stage emerges from the snail, typically encysts on vegetation and becomes the infective stage to be ingested by herbivores. Cattle, goats, sheep and camelids may ingest the parasite which migrates from the intestine to the liver where significant liver damage may occur due to migration of the fluke within the liver parenchyma. Infected species other than deer are considered dead end hosts since the full life cycle of the parasite will not occur in these species, therefore eggs of the fluke are not passed in the feces of the dead end hosts, limiting the ability to diagnose the presence of the parasite in these animals.

Two alpacas from the same farm in Franklin County have recently been diagnosed with **Fascioloides magna** at the Rollins laboratory. A third alpaca from the same farm was submitted for necropsy. While flukes were not identified in the liver of that animal grossly, histologic examination indicates hepatic damage secondary to fluke migration. **Fascioloides magna** is suspected in the third alpaca.
Increasing deer populations, unusual seasonal weather and/or possible range changes of this fluke are some factors to consider in North Carolina but have not been concluded. Pictures of diseased liver in the miniature llama are noted below. Note the adult fluke that was easily expressed from the surface of the liver in the first picture and the diseased liver on the cut surface from the migrating flukes in the second picture.

Dr. Reg Ridenhour and Dr. Mahogany Caesar

Equine

A 18 year old grey colored female Thoroughbred was presented to NCVDLS for post mortem examination. The provided history stated the horse had developed signs of colic and a large firm mass was palpated behind the spleen. There was blood seen on an abdominal tap, then the horse developed diarrhea, anorexia, tachycardia and a fever.

A summary of the post mortem examinations findings included: over 10 liters of bloody fluid in the abdominal cavity, infiltrative black colored tumors throughout the liver, a greater than 20 cm wide hematoma around the left kidney, a 15 cm plus wide black colored solid tumor adjacent to the left kidney along the dorsal midline with smaller black colored masses were present throughout the body and the spleen was massively enlarged with raised, nodular masses throughout the organ.

Histopathologic examination of the tissues were diagnostic for metastatic melanomas in the liver, intestine, lymph nodes and spleen, with an acute perirenal retroperitoneal hematoma.

It is estimated that more than 80% of gray horses over the age of 15 will develop at least one melanoma
tumor during its lifetime. Melanomas are tumors of the melanocytes, the cells that produce skin pigment. There appears to be a disturbance in the metabolism of melanin in aging gray horses, which stimulates local over-production of dermal pigment. Why gray horses are particularly susceptible isn't yet clear. Unlike melanomas in humans, which might be triggered by overexposure to ultraviolet radiation, gray-horse melanomas don't appear to be linked to an overdose of sun. The shady locations in which they tend to develop are well-protected from U/V radiation.

Melanomas can be hard or soft, solitary or appearing in clusters. Often, they're subcutaneous when they first appear, covered by normal skin, but as they develop over time, they become more obvious, and their surfaces can become ulcerated and/or infected. Although they're generally dark brown, gray, or black in color, it's also possible for these tumors to be unpigmented (amelanotic), a situation which makes diagnosis considerably more challenging.

The underside of the tail, the perineal and peri-anal regions, and the penis and sheath in males are the most common locations for melanomas. They also can be found on the ear margins, elsewhere on the head, in the jugular region, and near or on the parotid salivary gland. Much of the time, melanomas are fairly benign, slow-growing tumors, and are more unsightly than dangerous. They might remain that way for years or even decades. Within each tumor, however, lurks the potential for change to malignant growth, which swiftly can change the situation from a cosmetic nuisance into something life-threatening.

Metastatic melanomas might appear as rapidly spreading series of lumps or nodules, or even as vast, rippling sheets of black tumor masses across the tissue. Malignant melanomas can interfere with a horse's excretory functions, with breeding and foaling, or, if found in other locations on the body, such as the back or neck, can interfere with working under saddle or in harness.

Because malignant melanomas frequently appear in multiple sites, and because they have a high rate of metastasis, they can be very difficult to cure. When melanomas spread internally they commonly spread to the serosal surfaces of the liver, spleen, and lungs.

Gray horses aren't the only ones affected; horses of other coat colors also can develop melanomas. The incidence in non-grays is much lower, but when melanomas do appear in these horses, the tumors are more likely to be aggressive.

Dr. David Drum
WADDL was contacted May of 2013 by an individual owning a pond which was stocked with 2 to 3 year old Bream (primarily Bluegill), Bass, and Catfish and in the preceding 2 to 3 month period he reported that, in the Bream only, he was seeing bulging of the eyes. No deaths had occurred. He also estimated that in that time period he had caught 20 to 30 of these fish and that they all seemed to be generally healthy and that “if a hook happens to stick in the fish’s eye a red thread comes out”.

(I said to myself “I’ve got to see this to believe it!” Then, I said the same thing to the client. And he obliged. In the meantime I also called a PhD fisheries/aquaculture specialist at the local research farm who said, to me, “I’ve got to see this to believe it!”)

Four live Bluegill fish in pond water were submitted for evaluation. The fish were observed at rest off and on over a 45 minute time period during which no abnormal behavior was noted prior to each fish being humanely euthanized via Finquel (tricaine methanesulfonate).

The case report: The carcasses are those of two female and two male Bluegill sunfish with each having a body weight of 120 grams. Each of the 4 fish has mild (barely discernible) bilateral exophthalmos. In each fish a dull red line can be visualized at the periphery (usually anterior/medial) of the eye. Excision of the eye allows for retrieval of one (sometimes two) tightly wound ball of reddish-gray tissue that, when teased out to length, is a 10 cm long by 0.5 mm wide live nematode (photo). Direct microscopic examination of the nematode reveals each one to be a gravid female that is in the process of disgorging myriad live microfilaria. Direct microscopic examination of external mucus scrapings is negative for ectoparasites. Low numbers of gill flukes (Dactylogyrus sp.) are observed on direct microscopic examination of gill preps. The systematic necropsy examinations are concluded with no additional significant gross tissue alterations encountered.

Histopathology: Sections of fish brain, eye, heart, gill, stomach, intestine, liver, kidney, ovary, and skeletal muscle were examined. Morphological Diagnosis: Eye; scleritis, moderate, chronic, heterophilic and granulomatous, regionally extensive, with intraleisional nematode. Peter Moisan, DVM, DACVP

Laboratory Diagnosis: Scleritis due to *Philometra spp. oculo-orbital nematode*
Philometra is the genus and intraocular is the presumed species – e.g. Philometra intraocular. This Filariidae nematode of Bluegill and Sunfish is typically seen in fish over 1 year of age. Copepods are the intermediate hosts. Fish consume the copepods containing the immature male and female worms which then mature and mate within the fish’s gastrointestinal tract after which the male dies and the female then migrates to the oculo-orbit and gives birth (via protrusion of live, motile microfilaria) into the environment. Localized tissue reaction is not uncommon.

Dr. Richard C. Oliver

DEPARTMENTAL NEWS

ROLLINS LABORATORY

Rollins Lab New Hires
Dr. James Trybus, Pathology Services Coordinator, June, 2014 (start date)
Dr. Kristen Crook, Serology Section Supervisor, December, 2013
Allie Wetzel, Necropsy Assistant, December, 2013

Rollins Lab Promotions
Dr. Chad Cecil, formerly Virology Section Supervisor, was promoted to Section Head of Virology, February, 2014

Rollins Lab Resignations
Dr. Peter Moisan, Pathologist, December, 2013

Rollins Lab Retirements
Mary Denise Parker, Virology Medical Laboratory Technician, December, 2013
Sharon Greer-Graham, Virology Medical Laboratory Technician, December, 2013

ELKIN LABORATORY

Elkin Lab New Hires
Dr. Jessica Kees, Veterinary Diagnostician, March, 2014 (start date)

Facility Update
New HVAC Chiller unit being installed currently at NWADDL.

MONROE LABORATORY

Monroe Lab New Hires
Todd Walters, Veterinary Laboratory Assistant II, February, 2014
CE ATTENDANCE

Dr. Jennifer Haugland was invited to present during the Aquatic Session at the North American Veterinary Conference in January 2014 in Orlando. One presentation, Fish Diagnostics for the Private Practitioner, discussed the different diagnostic tests that can be done with sick fish. The other presentation, Help! Why are my koi dying?, discussed a diagnostic case of koi herpes virus and the available diagnostic tests.


Dr. Karen Post, Section Head of Bacteriology and Bacterial Serology (also Director of Laboratories), was honored at the annual conference with the AAVLD Award for Excellence in Diagnostic Veterinary Microbiology. Supported by Biomic Inc., the award for excellence in diagnostic veterinary microbiology (bacteriology) recognizes individuals for contributions and accomplishments in their field that result in new findings that have application for the betterment of diagnostic veterinary medicine.

Dr. David Drum attended a meeting on “Veterinary Biomedical and Diagnostics Sciences” at the University of Tennessee in December, 2013.

Drs. Mary Swanson and David Drum attended the Southeastern Goat and Sheep Seminar in Piedmont, SC in November 2013.
Directory

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Director
  Dr. Karen Post
Assistant Director
  Dr. Richard Mock
Veterinary Pathologists
  Dr. Tahseen Abdul-Aziz (Avian)
  Dr. Steven Rushton (Anatomic)
  Dr. Alison Tucker (Anatomic)
Veterinary Diagnosticians
  Dr. Jennifer Haugland
  Dr. Stacy Robinson
  Dr. Mahogany Caesar
Veterinary Microbiologists
  Dr. Karen Post
  Dr. Chad Cecil
  Dr. Richard Mock
Laboratory Section Supervisors
  Vacant — Virology
  Sandy Murphy — Bacteriology
  Mary Baker — Histopathology
  Dr. Kristen Crook — Serology
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Diagnostic Laboratory Advisory Committee

Dr. Allen Cannedy  Small Ruminant/Camelid Practitioner
Dr. Eric Gonder  Corporate Poultry Practitioner — Goldsboro Milling
Dr. Jennifer Haugland  Veterinary Diagnostician — NCDA&CS Veterinary Diagnostic Laboratory System
Dr. Shannon Jennings  Corporate Poultry Practitioner — Nash Johnson Farms
Dr. Randy Jones  Private Veterinary Practitioner — Livestock Veterinary Services
Dr. Richard Kirkman  Private Veterinary Practitioner — Large Animal
Dr. David Marshall  State Veterinarian — NCDA&CS Veterinary Division
Dr. Karen Post  Director of Laboratories — NCDA&CS Veterinary Diagnostic Laboratory System
Dr. Rick Sharpton  Corporate Poultry Practitioner — Perdue, Inc.
Dr. Betsy Sigmon  Small Animal Practitioner — Creature Comforts Animal Hospital
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