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VIRGINIA ANIMAL DIAGNOSTIC NEWSLETTER

A joint publication between Virginia Department of Agriculture and Consumer Services and the Virginia Tech Animal Laboratory Services

EDITORIAL

“From the Field to the Lab...”

During my first year of veterinary school, a professor commented about a veterinarian she admired who was the “best diagnostician she’d ever known.” I took that to mean someone who could get to the bottom of a case and get the answer, and I decided then that I wanted to aspire to that as well. At the time, I was headed toward mixed general practice close to my hometown. Specializing in pathology greatly interested me, but family commitments made pursuing a residency impractical. When the job opening was posted for the Lynchburg Regional Animal Health Laboratory Veterinary Diagnostician, I saw a niche that would let me apply my field experience to the elements of gross pathology and diagnostics I loved the most.

Although I was unsure of my qualifications when I applied, it turns out that my field experience was a great asset to becoming a diagnostician. Performing necropsies for the agriculture community is the primary role. It required someone who could directly communicate with clients and interpret complicated results directly for the submitter. General practice veterinarians do this every day, and I had plenty of practice getting this right (and many times wrong) before starting in this role. It also gave me considerable exposure to production settings by being on farms, seeing what farmers did, and talking to them. This has been crucial in drawing out historical information that is often important to reaching a final diagnosis.

There was, and is, plenty of learning to do as well. Although all veterinary students receive training on necropsies as part of the core curriculum, most general practitioners do not perform many after graduation (except some strictly food animal vets). Refresher training at the three other regional laboratories, along with several days back at Virginia-Maryland, my alma mater, was immensely helpful. It refined my necropsy

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Virginia-Maryland
College of **Veterinary Medicine**



VIRGINIA DEPARTMENT
OF AGRICULTURE AND
CONSUMER SERVICES

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skills and connected me to a network of great colleagues who were willing to answer questions and stay in touch as I started my new role. Skills such as trimming samples for histology and documenting forensic cases were brand new to me, and, though challenging at first, have quickly become part of my routine work. Taking on this role has been a wonderful fit for me, and I am excited to continue learning and growing in this career.

Taylor Young, DVM
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Horses and Donkeys



A rare malignant tumor in a donkey

A 20-year-old donkey stallion was evaluated due to a history of hyporexia, weight loss, and ADR. Ultrasonographic examination revealed large amounts of free fluid in the thoracic and abdominal cavities and a presumptive diagnosis of neoplasia was made. The animal was euthanized and submitted for necropsy. Postmortem examination revealed greater than 75 liters of red-tinged turbid watery fluid in the peritoneal and pleural cavities. Multiple lymph nodes were enlarged and effaced by pale gray neoplastic masses. Numerous (greater than 100) 1-5mm diameter firm gray nodules, arranged in linear strands that resembled rosary beads and tracked vessels, were disseminated throughout the mesentery, mesocolon, omentum, and serosal surfaces of the abdominal viscera. Based on histological examination, the diagnosis was lymphangiosarcoma. This is a rare malignant neoplasm arising from lymphatic endothelium. Among domestic animal species, lymphangiosarcoma is most typically seen in dogs and cats; reports in equids are rare (Vet Pathol 39:266-268 (2002)).

Thomas Cecere DVM, PhD, DACVP - Virginia Tech

Rabbits and Wildlife



Cecal round cell tumor and segmental hemorrhagic enterotyphlitis in a rabbit

A 7-year-old female spayed domestic rabbit was submitted for necropsy, with a history of strange behavior, hyperventilating, lethargy, and not eating properly. During the autopsy, a segmental hemorrhagic enteritis/typhlitis was noted in the distal jejunum and cecum, with the formation of a thick pseudo membrane in the lumen and numerous intraluminal Gram-positive bacillary bacteria. Also in the cecum, an infiltrative mass composed of atypical round cells was expanding the cecal wall. The final diagnosis was an enteropathy with a primary infiltrative mass in the cecum and secondary bacterial infection/ overgrowth, with Gram-positive bacilli. Pseudomembranous enteritis in rabbits is typically associated with toxigenic clostridial infections, most commonly *Clostridium spiroforme*. In this patient, it is presumed that the cecal tumor led to bacterial proliferation, toxin production, and, ultimately, severe enterotyphlitis.

Alexandra Reddy DVM - Virginia Tech
Francisco R Carvalho MV, DSc, DACVP - Virginia Tech

Encephalitozoonosis in an adult rabbit

A 2-year-old, female spayed rabbit with a history of intermittent dragging of the hindlimbs, urine scald, and anorexia was received for postmortem examination. At necropsy, the lungs were red and soft, and the thorax, pericardium and abdomen displayed increased amounts of fluid. Histologically, a multi-systemic inflammatory process was identified, involving the brain, spinal cord, kidney, liver, and heart, with numerous intralésional protozoal structures. These structures stained positive with Gram stain, which confirmed an infection with *Encephalitozoon cuniculi*. This organism is an obligate intracellular microsporidian parasite that is widespread in domestic rabbits. Although usually subclinical, heavy infestations of this parasite can lead to nervous system disease, leading to ataxia, paralysis, and muscular weakness, consistent with this patient's clinical history. Transmission in adult rabbits typically occurs through ingestion of spore-contaminated urine, followed by hematogenous spread to multiple organs, most commonly the brain, kidneys, and eyes, as well as other organs, including the liver, heart, and lung, which occurred in this case. Antemortem diagnosis is challenging, and definitive diagnosis is typically achieved by histopathology.

Alexandra Reddy DVM - Virginia Tech
Francisco R Carvalho MV, DSc, DACVP - Virginia Tech

Parvovirus in raccoons

A male and female raccoon, both approximately 8 weeks old, were submitted for necropsy by a wildlife rehabilitator. The rehab facility, as well as several other local rehabs, had been experiencing high morbidity and mortality in their raccoon patients. Inappetence, diarrhea, and lethargy were observed in both animals for approximately 5 days before death. The male tested positive for canine parvovirus on a SNAP test antemortem, but the female tested negative.

At necropsy, the male had mucoid, bile-tinged stomach contents with a small amount of milk and a small amount of tan, liquid feces in the colon. The female had a large amount of clotted milk in the stomach, with watery yellow intestinal contents in the ileum and colon. The

necropsies were otherwise unremarkable. Histopathology identified severe necrotizing and erosive enteritis in both animals, with the female showing additional damage to the liver and kidney suggestive of sepsis. The virus was detected on Canine parvovirus RT-PCR run on the intestine from the female, and culture and susceptibility from her kidney and liver revealed *E. coli* that was resistant to multiple antibiotics.

Canine parvovirus, as well as feline parvovirus (feline panleukopenia), are a common cause of disease of young raccoons in the rehabilitation setting and can cause high mortality as seen here. The timing of testing can complicate diagnosis, as viral shedding may not correlate with the onset of clinical signs. Prevention focuses on strict disinfection, quarantine, and vaccination protocols. There is no direct treatment for the virus, so treatment is supportive in nature to combat dehydration and nutrient losses and identify and treat secondary complications such as bacterial infections like was seen here.

Taylor Young, DVM - VDACS Lynchburg RAHL

Avian



Marek's disease in a chicken

A one-year-old Frizzle hen was submitted for necropsy with a history of weight loss, anorexia, and self-isolation. On necropsy, hundreds of tiny white nodules coated the mesentery and ovary, and the liver was enlarged with multifocal areas of pallor. Histopathology identified neoplastic lymphocytes within multiple organs and the sciatic nerves. A diagnosis of lymphoma was made with Marek's Disease as the most likely cause. The owner requested further testing and immunohistochemistry, and a Marek's

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Disease PCR was performed. T-cell markers were identified on neoplastic lymphocytes in the liver and spleen, and wild-type Marek's disease virus DNA was found in the liver. With this information, a definitive diagnosis of lymphoma due to Marek's disease was made.

Marek's disease is caused by an avian-specific herpesvirus found worldwide, which causes an untreatable and fatal disease. Before developing effective vaccines, this disease stymied large-scale chicken farming and remained a top cause of death in small flocks. Definitive diagnosis is difficult, especially prior to death. Most unvaccinated chickens eventually become infected, but only some develop the illness. Because of this, most would test positive for the virus even if they have not developed the disease. Definitive diagnosis can be made after death by identifying the virus within diseased tissues, although a presumptive diagnosis is usually sufficient. Purchasing vaccinated chicks or vaccinating newly hatched chicks is the best means of control.

Hailey Quercia DVM, MS, DACPV - Harrisonburg RAHL

Avian encephalomyelitis

A 4-week-old Ogye chick was submitted for necropsy. The chick had a sudden onset of an inability to stand but continued eating and drinking for two weeks before dying. Seven other chicks in the same brooder were affected similarly. Gross necropsy was unremarkable. Histopathology identified perivascular cuffing of plasma cells and lymphocytes in the brain, with rare glial nodules and sporadic central chromatolysis of neurons. There were nodular lymphocyte proliferations in multiple organs, as well as focal granulomas in the bursa and lungs. Avian encephalomyelitis was diagnosed presumptively. Avian encephalomyelitis is a viral disease of poultry that affects chicks less than 3 weeks old with signs similar to this chick. It can be spread vertically (parent to offspring) or horizontally (chicken to chicken). Commercial vaccinations are effective and available, and immunity acquired through natural exposure is lifelong without continued viral shedding.

Taylor Young, DVM - VDACS Lynchburg RAHL

Incubation issues in turkey poults

Ten four-day-old turkey poults were received for necropsy examination, with a history of multiple deaths in the flock. On arrival, some appeared weak and unable to stand, and many had primary flight feathers extending outward away from the body. All the poults were of different sizes. All had dark material stuck to the umbilicus, and four had overly large yolk sacs within the body.

All findings could be linked to issues in incubation. Maintaining ideal temperature and humidity during incubation is critical for proper development. These issues often cause underutilization of nutrients from the yolk sac during development, leading to variation in size and underdeveloped intestinal tracts, hearts, and skeletal muscle. It has been suggested as a cause of abnormal wing development. In these birds, the yolk sacs are large, leading to difficulties during absorption by the bird prior to hatch. This can prevent the umbilicus from fully closing, providing an opening for bacterial infection, a leading cause of death in young poultry. This case underscores the need for accurate monitoring of incubation parameters to promote the best chance of success for newly hatched birds.

Hailey Quercia DVM, MS, DACPV - Harrisonburg RAHL

Companion Animals



Pulmonary contusion and bulla associated with blunt thoracic trauma in a dog

A 4-month-old male Siberian Husky was hit by a car and died 72 hours later. The initial clinical exam, performed 24 hours after the accident, showed skin laceration over the right carpus. No evidence of internal bleeding was identified, and thoracic X-rays showed no

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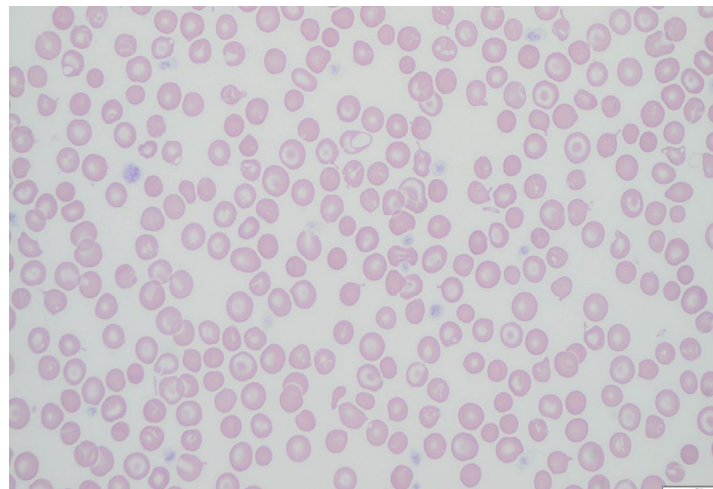
abnormalities in the thorax. Approximately 72 hours later, the animal died unexpectedly and was submitted for postmortem examination. Necropsy revealed a 4.5 x 2.5 x 0.5 cm subpleural bulla in the dorsal aspect of the right caudal lung, and filled with abundant air and scant blood. The remainder of the right lung was meaty and mottled dark red. Overall, gross and microscopic lesions were compatible with a pulmonary contusion associated with blunt thoracic trauma. Blunt trauma results in the transfer of kinetic energy to the thoracic wall, causing lung compression and overexpansion, leading to alveolar disruption, increased capillary permeability, fluid leakage into the alveoli, reduced lung surfactant, alveolar collapse, and impaired ventilation. In this case, there were no external injuries in the thoracic wall, probably due to the increased elasticity of the thoracic wall in young dogs.

Santiago Diab DVM, DACVP - Virginia Tech

Portosystemic shunt mimicking iron deficiency in an adult dog

An 8-year-old, neutered male shih tzu presented to the veterinary teaching hospital for a year-long history of intermittent episodes of collapse/possible seizure activity. A CBC and serum biochemistry were performed. The CBC revealed microcytosis (MCV: 42.6 fL, RI: 61.6-73.5 fL) and hypochromasia (MCHC: 24.9 g/dL, RI: 32.0-37.9 g/dL), though there was no anemia (PCV: 38.8%, RI: 37.0-62.0%). A review of the blood smear confirmed the microcytosis and hypochromasia, but also showed moderate to marked poikilocytosis due to target cells, keratocytes, and schistocytes (see photo). Based on these findings, an interpretation of suspected iron deficiency was made.

The serum biochemistry revealed multiple decreased indirect liver function markers (BUN: 5 mg/dL, RI: 9-30 mg/dL; Albumin: 2.4 g/dL, RI: 2.8-3.7 g/dL; total bilirubin 0.1 mg/dL, RI: 0.2-0.4 mg/dL; and cholesterol: 100 mg/dL, RI: 129-332 mg/dL) and a mildly elevated ALT (ALT: 80 U/L, RI: 16-75 U/L). Upon evaluation of the serum biochemistry and discussion with the clinician, a portosystemic shunt was suspected rather than true iron deficiency.



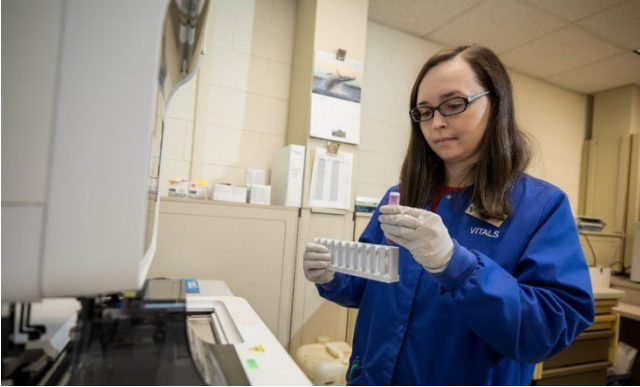
Peripheral blood smear, Modified Wright stain, 100x magnification: Many of the erythrocytes are microcytic and/or hypochromic. There are also numerous target cells, keratocytes, and schistocytes, which are common findings in iron deficiency anemia.

Fasting and postprandial bile acids were elevated (fasting BA: 128.5, RI 0.0-14.9; postprandial BA: 60.3, RI: 0.0-29.9) were consistent with decreased hepatic function. An abdominal ultrasound revealed a liver that was mildly reduced in size, but assessment of the portal blood flow was impossible due to the patient's conformation. A presumptive diagnosis of portosystemic shunt was made, and the patient was discharged with a treatment of lactulose, Denamarin and Keppra.

This case serves as an important reminder to consider liver disease/portosystemic shunt as a differential for microcytic and hypochromic anemia. The exact mechanism of the anemia is not known but is suspected to be due to decreased production of proteins needed for iron transport to erythrocyte precursors rather than a true iron deficiency. In addition to iron deficiency and liver disease/portosystemic shunt, other considerations for microcytic hypochromic anemias include copper deficiency and vitamin B6 deficiency.

Natalia Strandberg, DVM, MS, DACVP (Clinical) - Virginia Tech

LABORATORY NEWS



VITALS

With the days getting shorter, and Virginia Tech back in session, we are starting to see signs of Fall. Be advised that when bringing cases to the lab, traffic will be heavier, especially on Fridays and weekends when Hokies Football is at home. We have two new faces in lab central receiving - Hailea Martin and Kimberly Winck have joined us in April and have been a fantastic addition to our team! Drs. Alex Reddy, Laura Quishpe and Jasmine Yeh joined us as our newest Anatomic and Clinical Pathology residents. We're excited to have them and look forward to all the fantastic things they will do. The microbiology laboratory has expanded the number of serology and parasitology tests - we now offer a *Brucella canis* antibody test, Equine Infections Anemia ELISA, Anaplasma Antibody ELISA, Bovine Leukemia Virus Antibody ELISA, and Mini-FLOTAC (fecal egg count). We will be renovating and expanding the microbiology lab to facilitate the test expansion. We continue to provide pathology and histology support to the VDACS lab, and value our partnership and close interactions. And, as an AAVLD accredited lab, we are focused on continuous improvement. If you have any concerns or suggestions on how we can better serve you, please do not hesitate to reach out. We'd love to hear from you!

Tanya LeRoith DVM, PhD, DACVP. VITALS Director

VDACS

Testing for Highly Pathogenic Avian Influenza Virus in milk has continued at the Harrisonburg Regional Animal Health Laboratory, with over 240 samples tested to date (all testing negative). Federal orders have restricted interstate movement, enhancing surveillance through pre-movement testing of lactating dairy cows. Testing has also continued for Avian Metapneumovirus, with over 6,600 samples tested to date. In Warrenton, analysts are successfully working on bringing in CL SHI testing capabilities and are continuing to maintain a heavy necropsy and CEM culture load. The Wytheville Laboratory is continuing the last testing protocols in its annual fish testing in support of Virginia's Department of Wildlife Resources and some limited private producers. This testing covers both diagnostic and regulatory testing for fish health in the Commonwealth and surrounding states. Necropsy workload has remained steady, and serology testing has remained heavy. The Lynchburg Laboratory has seen a steady volume of necropsies after resuming this service in November 2023 and has been maintaining a steady workload regarding to PCR and Food Safety testing. In FY2024, over 5,000 samples were tested in the Food Safety Laboratory in support of private and commercialized producers.

Initial schematic design plans for the expansion of Warrenton, Harrisonburg, and Lynchburg Regional Animal Health Laboratories were submitted to the state's Department of Engineering and Budget for review and approval. A total of 8,800 sq feet will be added between the three laboratories.

VDACS OLS hosted the biennial audit and maintained accreditation of all four Regional Animal Health Laboratories (RAHL) from the American Association for Laboratory Accreditation (A2LA) through 2026, and brought all National Animal Health Laboratory Network (NAHLN) testing onto the A2LA scope for accreditation. Individual Standard Operating Procedures (SOPs) for the NAHLN testing performed at multiple sites were combined into one system SOP instead of having duplicate SOPs based on location. Testing additions and changes bring the total accredited testing methods up to 17 for Harrisonburg, 19 individual tests, and one

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panel consisting of 8 pathogens for Lynchburg, 7 for Warrenton, and 15 for Wytheville, comprising 71 SOPs. These testing methods and additional SOPs must maintain all the requirements of the ISO 17025:2017 standard, requiring continued monitoring via our Quality Management System, and are subject to rigorous internal audits to ensure the quality standards are met.

New hires in the system include a new administrative and laboratory assistant in Warrenton- Barbara Rocha, and a new serologist in Harrisonburg- Allison Sanford. An internal transition occurred with Gillian Slekar moving into the Harrisonburg Dairy Microbiologist and Laboratory Evaluation Officer position.

We are always open to feedback and ideas for testing to further assist our agricultural community. It is our goal to make this lab system the best it can be to further serve our clients. Our partnership with the Virginia Tech Animal Laboratory Services Lab (ViTALS) continues to evaluate ways to collaborate and expand testing services. Whether by utilizing data for reports or enhancing testing capabilities, we look forward to continuing to work together and with all of you.

Jessica Walters DVM, PhD, DACPV. Program Manager, Office of Laboratory Services

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