2025 Late Breaking Posters

2: EVALUATION OF PLASMA SYMMETRIC DIMETHYLARGININE AS A BIOMARKER FOR BOVINE KIDNEY DISEASE

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Background: The efficacy of serum urea and creatinine in the diagnosis of bovine kidney disease is limited because of their low sensitivity and specificity. Serum symmetric dimethylarginine (SDMA) concentration is correlated with renal glomerular filtration rate and has been reported to be useful for the diagnosis of kidney disease in other species.

Hypothesis/Objectives: To test the hypothesis that SDMA may be an effective biomarker for diagnosing kidney disease in cattle. The objectives were to assess the analytical performance of SDMA measurement using a bench-top analyzer, to establish an SDMA reference interval in cattle, and evaluate the efficacy of plasma SDMA for the diagnosis of bovine kidney disease.

Animals: 199 healthy animals (53 male and 96 female beef cattle and 50 female dairy cows) and 27 azotemic cows with confirmed kidney disease

Methods: A partial analytical validation of SDMA measurement with bovine plasma was performed including assessments of short- and long-term imprecision, linearity, and stability. The reference interval was established following ASVCP recommendations. Finally, the diagnostic efficiency of SDMA was assessed by the determination of specificity, sensitivity, and the AUC of the ROC curve

Results: SDMA measurements showed acceptable analytical performance. The reference interval was 60–160 µg/L. There was no significant difference according to sex, type of breed and food supply. Using the upper limit of the reference interval (160 µg/L) as the cut-off, sensitivity and specificity were 0.85 and 0.98, respectively.

Conclusions: SDMA at the threshold of 160 µg/L is an effective marker for diagnosing kidney disease in cattle.

3: CANINE COPPER TOXICOSIS ASSOCIATED WITH ACUTE RENAL FAILURE IN A BOXER-MIX WITH A HOMOZYGOUS ATP7B VARIANT

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A 7-year-old spayed female Boxer-mix presented to the NCSU Small Animal Emergency Service for severe oliguric and suspected acute kidney injury following acute liver injury. Initial diagnostics revealed significantly high ALT, severe azotemia (IRIS Grade V), hypophosphatemia, prolonged PT/PhTT, and paradoxical glucosuria. The dog progressively became anuric and overhydrated despite being on continuous rate infusions of furosemide and diltiazem, and euthanasia was elected. Gross necropsy findings included pulmonary edema, thoracic effusion, and prominent pale striations in the renal cortices. The liver appeared grossly normal. Histologic evaluation revealed moderate subacute centrilobular hepatocellular necrosis and diffuse, severe copper accumulation (Grade IV/V) demonstrated by rhodanine staining. The kidney had severe acute multifocal proximal tubular necrosis and karyomegaly with intracytoplasmic copper granules. An acquired Fanconi-like syndrome was suspected based on the renal microscopic changes and clinical findings of marked azotemia and hypophosphatemia despite significant oliguria. Quantitative analysis demonstrated high liver (2,625 ppm dry weight; RI 105–350) and kidney (199.5 ppm dry weight; RI 17.5–52.5) copper concentrations consistent with toxicity. Genetic testing revealed homozygosity for the ATP7B variant associated with copper toxicosis in dogs. The hepatic copper accumulation associated with ATP7B variant homozygosity, coupled with subacute hepatocellular necrosis and acute renal tubular necrosis, supports the release of stored copper from the liver, resulting in accumulation and damage to proximal renal tubules. However, the inciting cause of the liver damage is unclear. This case demonstrates the potential for copper toxicosis in a non-breed predisposed canine and the potential for increased risk of acute renal injury.

4: DETECTION OF BILE CASTS IN CIRCULATING LEUKOCYTES IN TWO DOGS

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Recently, inclusions within circulating leukocytes have been described in dogs. We describe two cases with bile casts inclusions.

Case 1 : A 6-year-old female neutered Staffordshire Bull Terrier was referred to the veterinary teaching hospital of Toulouse (ENVT) in France for a suspicion of IMHA. The dog exhibited lethargy, dysorexia, pigmenturia, and icterus. Ultrasound showed severe cholecystitis. Biochemistry revealed hypoalbuminemia, marked increased ALP and GGT activities and severe hyperbilirubinemia. Haematology revealed severe macrocytic hypochromic regenerative anaemia, neutrophilic leukocytosis,left shift and toxic neutrophils. A positive Coombs test and spherocytosis confirmed IMHA. The dog died from cardiorespiratory arrest.

Case 2 : A 4-year-old female neutered Miniature American Shepherd was referred to the intensive care unit of ENVT for post-surgical management following stent placement on the duodenal papilla and duodenotomy, in a context of high-grade tubulo-trabecular carcinoma of exocrine pancreas. during hospitalization, ultrasound suggested severe cholecystitis, and biochemistry showed hypoalbuminemia and increase ALT and ALP activities. Haematology revealed moderate normocytic hypochromic regenerative anaemia, thrombocytopenia, left shift and toxic neutrophils. Following clinical deterioration, the dog was humanely euthanized.

In both cases, rare cylindrical, dark inclusions were observed on blood smears, in macrophages (Cases 1 and 2) neutrophils (Case 1). These inclusions were positive to Hall's bilirubin staining, confirming them as bile casts.

To our knowledge, these are the first two reported cases of bile casts in circulating leukocytes in dogs, confirmed by specific staining. The presence of these inclusions is most probably related to a severe cholestasis secondary to cholecystitis.

5: PRIMARY INTRACRANIAL ANGIOCENTRIC LYMPHOMA IN A DOG

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A 9.5-year-old spayed female Greyhound was presented with a 10-day history of a wobbly gait and vision loss in the left eye. Neurologic examination revealed appropriate mentation and ambulatory status with a hypermetric gait. As clinical signs progressed, the patient became obtunded and non-ambulatory tetraparetic by the following day, with marked postural deficits in all four limbs, most notably in the left forelimb. The patient remained blind in the left eye, with absent physiologic nystagmus. No lymphadenopathy or organomegaly was detected on physical examination or abdominal ultrasound. Magnetic resonance imaging revealed a large mass in the right parietal cerebral hemisphere, with extensive vasogenic edema and signs of increased intracranial pressure, including cerebellar compression and caudal transtentorial and subfalcine herniation. A right parieto-temporal craniectomy was performed, revealing an unencapsulated mass with poorly defined borders; the mass was partially debulked. Cytology showed a highly cellular sample composed predominantly of a monotonous population of intermediate to large lymphocytes with frequent mitotic figures, consistent with lymphoma. Histopathology showed an infiltrative neoplasm composed of sheets of intermediate to large lymphocytes displaying frequent angiocentrism and angioinvasion, with abundant central necrosis. Immunohistochemistry demonstrated CD20 immunoreactivity of neoplastic cells, surrounded by a reactive population of CD3-positive small lymphocytes. We report a case of primary B-cell lymphoma affecting the brain of a dog, an uncommon diagnosis. In addition, this report documents angiocentrism and angioinvasion in a pattern inconsistent with classic intravascular lymphoma but resembling the human lymphoid disorder known as lymphomatoid granulomatosis.

6: A FLAMMULATED OWL (PSILOSCOPS FLAMMEOLUS) MASS MORTALITY EVENT INVESTIGATION

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Introduction: Flammulated Owls (FLOWs) are small migratory raptors with dark eyes and flame-like streaks on the shoulders and back. AZ Game and Fish Department classifies them as Tier 2 Species of Greatest Conservation Need. In fall 2024, Maricopa Co wildlife rehabilitation centers admitted 54 FLOWs, compared to 3 in 2022 and 9 in 2023, with 44 deaths. This surge coincided with a heat wave in the Sonoran Desert—temperatures reached a maximum average of 102.2°F (81-117°F), potentially contributing to mortality during migration. We hypothesize that high temperatures caused increased mortality with infectious disease and toxicosis as possible contributing factors.

Methods: Necropsies of 41 owls (31-60.5g) (28 male (17 immature, 11 mature), 13 female (10 immature, 3 mature)) and histopathology.

Results: Gross exams revealed cloacal and fecal urates (n=26), ureteral urates (n=23), white renal stippling (n=23), and visceral gout (n=1). Some specimens had other lesions, mainly trauma-related, but these were inconsistent across the cohort and less significant. Fat deposit scores (graded 0-4) yielded a median of 3 (2.7 average). Renal histopathology detected acute tubular damage in 39 FLOWs.

Conclusion: Preliminary findings suggest most FLOWs developed fatal acute tubular necrosis due to dehydration. Starvation was an unlikely factor. Ongoing analyses include histopathology, diet composition via metabarcoding of gastrointestinal contents, infectious disease testing, and GC-MS, LC-MS/MS, and ICP-MS of organs to detect synthetic toxins, industrial chemicals, and heavy metals. This study will establish baseline data on environmental contaminants, disease, and diet in FLOWs and elucidate the impact of elevated temperatures on their health.

7: METASTATIC HEMANGIOSARCOMA IN 5 HORSES

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Hemangiosarcoma is an uncommon malignant neoplasm in horses. Here we describe 5 cases of metastatic hemangiosarcoma diagnosed in horses during postmortem examination at the University of Tennessee College of Veterinary Medicine. Presenting complaints included colic, lethargy, and anorexia (3/5), weight loss (1/5), and distress and ataxia (1/5). Hemangiosarcoma was the cause of clinical decline leading to euthanasia in 4/5, and complications related to treatment of anemia, presumptively due to hemangiosarcoma, was the cause of death in 1/5. The average time from initial

presentation to death or euthanasia was 11.4 days. Affected organs included the spleen (4/5), liver (3/5), adrenal gland (3/5), lung (2/5), kidney (2/5), adipose (1/5), brain (1/5), bone marrow (1/5), colon (1/5), diaphragm (1/5), and skeletal muscle (1/5). Unique histologic features included a thick fibrous capsule or abundant fibrous tumor stroma (5/5), large paucicellular regions (4/5), and regions with bland morphology of neoplastic cells (4/5). A majority of cases occurred in middle aged to older horses (average age 17.8 years), and no breed or sex predilections were identified. Hemangiosarcoma in horses may have a bland or paucicellular histologic appearance that can vary by location even in the same horse, and these features, in combination with a thick fibrous capsule and/or stroma, may result in a misdiagnosis of hematoma or hemangioma. These initial diagnoses in a horse may warrant further investigation for hemangiosarcoma.

8: ARTIFICIAL INTELLIGENCE- BASED IMAGE ANALYSIS OF NODAL CUTANEOUS MAST CELL TUMOR METASTASIS

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Canine cutaneous mast cell tumors (cMCTs) account for approximately 21% of all skin tumors in dogs in the US (Bostock). Lymph node metastasis is a negative prognostic indicator. Nodal metastasis is often graded via the Weishaar classification system (Weishaar et al). However, this grading system is unvalidated and unstandardized. A more objective and focused assessment may improve prognostication in dogs with nodal MCT metastasis. This study aims to use artificial intelligence-based image analysis to quantitate the morphological and molecular features in dogs with nodal cMCT metastasis. We have trained a model to quantify the nodal mast cell burden (total cKIT+ cells and density) and the proliferation quotient (cKIT+/ki67+). We believe that our model will identify criteria that will correlate with long-term survival in dogs with cMCT nodal metastasis, which will help to improve clinicians' decision making and therapeutic planning.

9: EVIDENCE OF CHRONIC WASTING DISEASE VERTICAL TRANSMISSION USING AMP-IHC: AN ENHANCED IMMUNOHISTOCHEMICAL METHOD FOR HIGH-SENSITIVITY PRION DETECTION

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Background and Objective: *Pasteurella* spp. are opportunistic bacteria commonly found in the oral cavity of animals and are recognized as zoonotic pathogens responsible for pasteurellosis. Pasteurellosis caused by *Pasteurella canis* is rare in both humans and animals. This study aimed to elucidate the pathological features of pasteurellosis in group-reared meerkats (*Suricata suricatta*; Ss) that developed pasteurellosis, and to investigate the infection status of *Pasteurella* spp. in Ss.

Results Diagnostic imaging revealed hydrothorax or pyothorax in four Ss. At necropsy, two Ss (10-month-old male and female) exhibited purulent pleural effusion in both thoracic cavities, along with pleural thickening, and pustulation. Histopathology revealed hypertrophy of serosa of lung and of thoracic wall characterized by lymphatic infiltration, mesothelial hyperplasia, organizing fibrin deposition, and pseudomembrane formation. *P. canis* was isolated from organs, including pleural effusion. *P. canis* was detected in all 20 Ss, and *P. multocida* was detected in 16 Ss in them.

10: VALIDATING IMMUNOHISTOCHEMISTRY ANTIBODIES FOR CANINE NODAL LYMPHOMA USING DIGITAL IMAGE ANALYSIS

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Background: Canine lymphoma is one of the most common malignancies in middle-aged dogs. Histology and immunohistochemistry (IHC) are considered the gold standard for diagnosing and subtyping canine nodal lymphoma. However, there are currently no standardized antibody panels recommended for IHC-based subtyping. Validation of IHC antibodies is therefore critical to ensure diagnostic accuracy, consistency, and standardization across laboratories.

Objective: To evaluate the performance of multiple B-cell and T-cell antibodies for immunohistochemistry (IHC) in subtyping canine nodal lymphoma.

Methods: Formalin-fixed, paraffin-embedded tissue blocks were prepared from canine lymph node biopsies with confirmed lymphoma and sectioned for hematoxylin and eosin (H&E) staining and immunohistochemistry (IHC). The IHC panel comprised of antibodies against CD79a, PAX5, CD20, CD5, and CD3. Nodal lymphomas were subtyped according to the canine-adapted World Health Organization (WHO) classification. IHC staining was quantitatively analyzed using HALO, a digital image analysis software program.

Results/Conclusion: A total of 24 lymphoma cases were evaluated, comprising of 12 B-cell and 12 T-cell lymphomas. Expected widespread staining with B- and T-cell antibodies was observed in 9/12 B-cell and 8/12 T-cell lymphoma cases. Several cases demonstrated aberrant immunophenotypic marker expression. Among the B-cell lymphomas, neoplastic cells lacked PAX5 expression in two cases, and aberrant cytoplasmic CD3 expression was observed in one case. Among the T-cell lymphomas, aberrant cytoplasmic CD20 expression was identified in four cases, two of which also lacked CD5 expression. These findings highlight the importance of validating a panel of B- and T-cell antibodies for immunohistochemistry to ensure accurate subtyping of canine nodal lymphoma.

11: WEAKLY SUPERVISED DEEP LEARNING MODEL FOR DETECTING CANINE CUTANEOUS MAST CELL TUMORS IN WHOLE SLIDE IMAGES

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Background: Accurate identification of mast cell tumors (MCTs) in canine skin biopsies is critical for diagnosis, grading for prognosis, and treatment planning. In computational pathology, many image-analysis machine learning models require labor-intensive, manual outlining of tumor boundaries for training. This process is time-consuming, requires expert input, and limits the scalability of such tools.

Objective: To develop and evaluate an attention-based multiple instance learning (MIL) framework for weakly supervised classification of whole-slide images (WSIs) as MCT-positive or non-MCT without requiring manual boundary annotations.

Methods: An in-house dataset of 485 canine WSIs was stratified into a development set (380 slides; 200 low-grade MCT, 60 high-grade MCT, 120 non-MCT/healthy) and a holdout set (105 slides; 45 low-grade MCT, 20 high-grade MCT, 40 non-MCT/healthy). Slides were tiled into 100 µm patches at 20×, resized to 224×224 px, and encoded using a pretrained Vision Transformer (ViT) to produce 384-dimensional feature vectors. The MIL network learned patch-level attention weights to aggregate features into slide-level embeddings for binary classification. Model selection was based on validation AUC.

Results: The model achieved high performance on both the validation set (AUC 0.953, F1 0.968, accuracy 0.943) and the holdout set (AUC 0.951, F1 0.964, accuracy 0.943). Generated attention maps highlighted regions consistent with tumor areas identified by pathologists.

Conclusion: This computational framework, based on weakly supervised MIL and deep learning, provides accurate MCT classification without regional annotations, while generating interpretable attention maps that may assist pathologist review. The approach has potential for scalable deployment in veterinary histopathology workflows.

12: MULTIPLE TUMORS IN AN AFRICAN PIGMY HEDGEHOG: A CASE REPORT

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4-year-old, male, African pygmy hedgehog presented with multiple lesions suggestive of neoplasia. A lesion was identified on the gingival mucosa, along with cutaneous lesions on the dorsal and abdominal regions. The biopsies were collected and fixed in buffered formalin for histopathological and immunohistochemical analysis.

The oral lesion exhibited moderate epithelial hyperplasia with mild loss of cellular polarity and an intact basement membrane. The dorsal cutaneous lesion consisted of a poorly demarcated, non-encapsulated neoplastic process, characterized by a predominance of multinucleated cells (exceeding 20 nuclei per cell) with marked pleomorphism and abundant cytoplasm, intermingled with spindle cells arranged in an orthogonal pattern. > 18 mitoses per 2.37 mm². The abdominal lesion was a poorly demarcated, non-encapsulated, multilobulated neoplasm composed of spindle cells arranged in orthogonal and parallel bundles. The neoplastic cells exhibited abundant, eosinophilic, fusiform cytoplasm, with round to oval nuclei containing granular chromatin and a centrally located prominent nucleolus. >16 mitoses per 2.37 mm². Both cutaneous lesions infiltrated subcutaneous adipose and muscle tissue.

One month after surgery, a new neoplastic mass appeared in the caudal region, and the patient died a few weeks later. The owners did not consent to a necropsy. Cases of histiocytic sarcoma have been reported in the spleen, intestines, skin, and as multiple concurrent tumors in this species.

13: POST-MORTEM FINDINGS IN AN OLIVE RIDLEY TURTLE (LEPIDOCHELYS OLIVACEA) STRANDED ON THE COAST OF CHILE

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An adult female olive ridley sea turtle (*Lepidochelys olivacea*) was received deceased after stranding at Los Pozos beach (coastal area of Huasco), within Llanos de Challe National Park, Atacama Region, Chile. Background information indicates the turtle stranded alive and was fatally attacked by feral dogs. Macroscopic examination revealed an open skull fracture involving the salt glands, bite-associated lesions on the right side of the carapace and pectoral fin, with erosive injuries and fissures. Moderate biofouling was observed with algae and barnacles on the carapace surface.

Necropsy findings included marked thickening of the esophageal, gastric, and intestinal mucosa, with severe edema, dilated lymphatic vessels, and congested blood vessels. Parasitic structures up to 8 mm were identified in the aortic lumen. Multiple tissue samples were fixed in 10% buffered formalin for histopathological analysis.

Microscopically, the skin of the skull, carapace, and flippers showed mild to moderate mononuclear inflammation, isolated heterophils, and hemorrhagic foci surrounded by connective tissue bands. Occasional plant structures were embedded in the dermis. In the gastrointestinal submucosa, multiple parasitic structures were observed in vascular and extravascular compartments, ranging from rounded to ovoid (up to 150 µm in length and 50 µm in width), with a thin cuticle. These were often surrounded by mononuclear inflammation, multinucleated giant cells, and collagenous fibroconnective tissue.

Severe gastrointestinal lesions could have contributed to the stranding event. The parasites are morphologically compatible with members of the superfamily Schistosomatoidea; however, molecular diagnostics are required for definitive identification.

14: GAS GLAND EPITHELIAL TUMORS WITH INTRATUMORAL GRANULOMAS IN TWO BLACK CRAPPIES

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Swim bladder tumors are rarely reported in teleosts but often arise from the gas gland, which is composed of a capillary rete mirabile and adjacent lactic acid secreting epithelial cells. Gas gland epithelial tumors are frequently classified as adenoma, papillary adenoma, or adenocarcinoma. In 2024, tissues fixed in 10% NBF from two aquarium-housed black crappies (Pomoxis nigromaculatus) were submitted to diagnostic services at North Carolina State University and University of Georgia Colleges of Veterinary Medicine. In case 1, the fish was negatively buoyant with a soft tissue opacity within the swim bladder lumen on radiographs. Grossly, the swim bladder was effaced by a brown to tan multilobulated mass which contained opaque, brown fluid. No clinical history was received for case 2. Histologically, both swim bladders were infiltrated by neoplastic epithelial cells which formed tubules or papillary projections with moderate cellular pleomorphism. Case 1 was consistent with a papillary adenocarcinoma based on the presence of mitotic figures and invasiveness of the neoplastic cells, and case 2 was consistent with a gas gland adenoma due to lack of mitotic figures. In both cases, chronic granulomas were within the swim bladder and tumor, with acid-fast positive bacilli within the granulomas in case 2. Case 2 additionally had a renal tubular adenoma with granulomas. Chronic inflammation may have promoted tumor development in both cases. To the authors' knowledge, these are the first reported gas gland tumors in black crappies, with one having malignant features which are rarely described in gas gland tumors.

15: HEMATOLOGY AND CLINICAL CHEMISTRY DATA FOR GERIATRIC AFRICAN GREEN MONKEYS (CHLOROCEBUS SABAEUS)

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Introduction: The African green monkey (AGM; *Chlorocebus sabaeus)* is a translational species that shares significant genetic, anatomic, physiologic, metabolic, and pharmacologic characteristics with humans and nonhuman primates (NHPs). Here, we present clinical pathology data from geriatric (>13 years old) AGMs and compare it to Virscio's in-house reference intervals (RIs) and published RIs for geriatric Caribbean AGMs.

Methods: The data represent geriatric AGMs from Virscio's colony on St. Kitts, collected between 2023-2025 from a mix of test article-naïve and off-study animals. All

animals were clinically healthy with a few having chronic health conditions not anticipated to affect clinical pathology parameters.

Results: Clinical pathology data were collected from 36 total geriatric AGMs. Seven AGMs were healthy, test article-naïve AGMs ranging from 13-16 years old (group 1). Sixteen were test article-naive, 17–28 year old AGMs with largely unremarkable clinical histories (group 2). Thirteen were from 15-27 years old with a mixed study history (group 3). Group 1 had no significant clinicopathologic dyscrasia compared to Virscio's RIs. The primary clinicopathologic abnormalities in Groups 2 and 3 were mildly elevated AST and ALT in 3 and 6 animals, respectively, compared to Virscio's adult AGM RI. These mild ALT elevations were within published RI for geriatric AGMs. Mild lymphopenia in some animals in this group was attributed to stress.

Conclusion/Impact statement: Hematology and clinical chemistry data for Virscio's geriatric AGMs aids in understanding the effects of age in this species, as well as interpreting pharmacology and nonclinical safety studies in a geriatric population.

16: BLUNT FORCE TRAUMA TO THE CHEST: COMPLETE RUPTURE OF CAUDAL VENA CAVA AND FATAL HEMOTHORAX IN A PUPPY

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Case History

A juvenile, 6.2kg, female, Shepherd mixed dog was submitted for forensic necropsy to Texas Tech School of Veterinary Medicine as part of a legal investigation. The dog reportedly collapsed during a flea bath and was presented deceased to a local veterinary clinic with a wet coat and frothy blood oozing from the nose, leading to the suspicion of drowning. The specific necropsy request was to determine the cause and manner of death.

Gross Findings

Gross examination revealed rare minimal to mild contusions on the trunk, pale mucous membranes, a completely ruptured caudal vena cava, extensive pulmonary and mediastinal lacerations, and a severe hemothorax (225 ml). Two consecutive left rib necks were focally enlarged by calluses. Preliminary histology of the lungs cannot confirm additional events of drowning.

Discussion

The discrepancy between the mild external contusions and the lethal internal injuries is attributed to the elasticity of the tissues in juvenile animals. The absence of foreign materials in the alveoli on histology does not completely exclude the possibility of additional drowning in particle-free water. Furthermore, the presence of rib calluses raises concerns about a previous episode of abuse.

Summary:

The cause of death of the dog was hypovolemic shock due to blunt force trauma to the chest, resulting in complete rupture of the caudal vena cava and massive hemothorax. The manner of death is classified as non-accidental killing, the equivalent to homicide in humans. An additional event of drowning cannot be confirmed or ruled out with certainty.

18: DIAGNOSTIC UTILITY OF CYTOKERATINS 7 AND 14 IN DIFFERENTIATING CANINE ANAL SAC AND PERIANAL CARCINOMAS

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Background: Perianal carcinomas (PACs)and apocrine gland anal sac adenocarcinomas (AGASACAs) are malignant perianal neoplasms with differing prognoses and treatments. Histopathological evaluation is typically sufficient for diagnosis. However, differentiating poorly differentiated PACs from solid-type AGASACAs can be challenging. Previous studies have identified cytokeratin (CK) 7 and CK14 immunohistochemistry (IHC) markers as having potential diagnostic utility when differentiating these neoplasm types.

Objective: To characterize CK7 and CK14 staining patterns of canine perianal carcinomas and AGASACAs and their potential diagnostic use.

Methods: CK7 and CK14 IHCs were performed on routine biopsy samples from 50 confirmed cases of PACs and AGASACAs submitted to the Iowa State University Department of Veterinary Pathology between 2015 and 2024. IHCs were performed according to previously published methods using commercially available antibodies. IHCs were assessed using a qualitative scoring scheme.

Results: CK14 IHC demonstrated positive staining in PACs and negative staining in AGASACAs. CK7 IHC was met with complications as the CK7 antibody clone OV-TL 12/30, used in previous publications, failed to stain any canine tissues. CK7 antibody clone EPR17078 produced cytoplasmic staining in AGASACAs with weak to negative staining in PACs.

Conclusions: Differentiating PACs from AGASACAs can be a diagnostic challenge, and incorrect interpretation is possible when samples are examined only by routinely stained sections. CK7 and CK14 IHCs exhibit diagnostic utility in differentiating these neoplasms, particularly when employed concurrently. However, careful selection of CK7 antibody clones and rigorous validation of the protocol are recommended.

19: PULMONARY ACINAR ADENOCARCINOMA IN AN ATRIAL MALIGNANT PARAGANGLIOMA: A CASE OF TUMOR-TO-TUMOR METASTASIS IN A FRENCH BULLDOG. MORA-IBAÑEZ MOLINA AND OLUFEMI O. FASINA DEPARTMENT OF VETERINARY PATHOLOGY, COLLEGE OF VETERINARY MEDICINE, IOWA STATE UNIVERSITY, AMES, IOWA, UNITED STATES.

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Tumor-to-tumor metastasis is a rare and unique feature associated with neoplasms in dogs and humans. Here, we report tumor-to-tumor metastasis in a 12-year-old female French bulldog that presented for consultation and was euthanized due to a right caudal pulmonary mass diagnosed via radiography. Necropsy revealed a firm, tan-white 4.5x2.5x3.5 cm mass within the right caudal lung lobe. In the heart, there was a firm 2 x1.5x 1.5cm nodular mass within the right atrium between the vena cava and pulmonary artery. Microscopically, the atrial mass revealed an infiltrative and densely cellular neoplasm composed of polygonal cells organized in nests and packets separated by a thin fibrovascular stroma. Neoplastic cells have round vesicular nuclei, 1-3 prominent nucleoli, moderate to abundant eosinophilic and granular cytoplasm, and indistinct cell borders. Within the right atrial mass, there is an infiltrative and densely cellular neoplasm composed of pulmonary polygonal cells organized in variably-sized acini filled with mucinous secretions and separated by thick scirrhous stroma. The alveolar spaces are expanded and effaced by similar neoplastic cells. The right atrial neoplastic cells lack chromogranin A expression and have nuclear and cytoplasmic expression of protein gene product 9.5 (PGP 9.5), and the pulmonary neoplastic cells have cytokeratin and nuclear thyroid transcription factor 1 (TTF1) expression. Intralesional TTF1-positive polygonal cells within the PGP 9.5-positive malignant paraganglioma cells are consistent with pulmonary acinar carcinoma metastasis to right atrial malignant paraganglioma, a tumor-to-tumor metastasis. To our knowledge, this is the first report of a tumor-to-tumor metastasis in an atrial malignant paraganglioma.

20: VIRULENCE OF EDWARDSIELLA PISCICIDA AND E. TARDA IN A MURINE MODEL OF INFECTION

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Edwardsiella piscicida is an emerging fish pathogen closely related and phenotypically indistinguishable from *E. tarda*, a known zoonotic bacterium associated with gastroenteritis and potentially fatal systemic disease in humans. Unlike *E. tarda*, there is no clear consensus on the risk *E. piscicida* poses to mammals. With increasing outbreaks of *E. piscicida* among popular food and sportfish, there is a crucial need to better understand the pathophysiology of edwardsiellosis in mammals and assess its foodborne/waterborne disease risk. In this study, C57BL/6, heterozygous (*nu/*+), and homozygous (*nu/nu*) mice were exposed to *E. piscicida* S11-285 or *E. tarda* 94-45-85 at a dose pathogenic to fish (~10^5 CFU/gram) via intragastric gavage or subcutaneous injection. Both immunocompetent (2/6 C57BL/6; 4/6 *nu/*+) and immunocompromised (2/6 *nu/*nu) mice injected with *E. tarda* developed lesions grossly similar to reported human *E. tarda* wound infections. Conversely, only a single immunocompromised mouse injected with *E. piscicida* and none of the orally inoculated mice developed clinical disease over the 21-day experiment. Histologically, *E. tarda* injected mice demonstrated suppurative inflammation with intralesional bacteria and regionally

extensive acute rhabdomyonecrosis at the injection site. Immunocompetent (*nu/+*) mice were significantly more susceptible to disease when injected with *E. tarda* compared to *E. piscicida* (One-tailed Fisher's exact test, p=0.0303), however there was no significant difference in the susceptibility of immunodeficient mice to *E. tarda* and *E. piscicida* (p=0.5). These findings suggest reduced pathogenicity of *E. piscicida* in immunocompetent mammalian hosts compared to *E. tarda* and that immunodeficiency could increase the risk of mammalian *E. piscicida* infection.

21: PHENOTYPIC CHARACTERIZATION OF COLLABORATIVE CROSS MOUSE LINE CC011/UNC

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The Collaborative Cross (CC) is a panel of inbred mouse strains, derived from eight founder lines developed for the genetic analysis of complex traits. Prior publications on the CC011/Unc line report a high incidence of colitis. Our unpublished observations include paronychia in females and fragile teeth in both sexes. Therefore, pathology phenotyping was undertaken to better characterize this line.

We evaluated tissues from five male and ten female 7-9 months-old CC011/Unc mice housed in the UNC Systems Genetics Core Facility (SGCF), both for gross and microscopic pathology. Blood collected at necropsy from 4 female and 5 male mice was analyzed. Tissues were fixed and examined through histopathology.

We identified several pathological outcomes in these animals. Suppurative paronychia and P3 osteolysis, mixed aortic root aortitis with dissection, and mixed folliculitis/furunculosis were evident only in females. In both sexes, ameloblast dysplasia, grossly evident as pearly white incisors. Mixed colitis, as previously described in both sexes. Both sexes had suppurative and necrotizing arteritis. Blood tests revealed that females had significantly higher white blood cell and neutrophil counts, indicating more widespread inflammation.

In summary, CC011 remains a strain with a variety of pathologies which are relevant to a number of human conditions. Female mice have unique inflammatory lesions in the heart and nailbeds, which have human immune-mediated disease correlates suggesting translational relevance. Further, nearly 100% incidence of ameloblast dysplasia in both sexes highlights this strain as a model for congenital enamel defects. These findings highlight the CC's potential for translational research.

22: PATHOLOGICAL FEATURES OF PASTEURELLOSIS CAUSED BY PASTEURELLA CANIS AND THE PREVALENCE OF PASTEURELLA SPP. IN MEERKATS, SURICATA SURICATTA

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Background and Objective: *Pasteurella* spp. are opportunistic bacteria commonly found in the oral cavity of animals and are recognized as zoonotic pathogens responsible for pasteurellosis. Pasteurellosis caused by *Pasteurella canis* is rare in both humans and animals. This study aimed to elucidate the pathological characteristics of pasteurellosis in group-reared meerkats (*Suricata suricatta*; Ss) that developed pasteurellosis, and to investigate the infection status of *Pasteurella* spp. in Ss.

Methods Four of the 25 Ss housed in the same facility developed pasteurellosis. All four were clinically examined, and two Ss underwent pathological and microbiological evaluations. Oral swabs were collected from 20 Ss (6–24 months old), including the two affected animals, to assess the presence of *Pasteurella* spp. using PCR.

Results Diagnostic imaging revealed hydrothorax or pyothorax in four Ss. At necropsy, two Ss (10-month-old male and female) exhibited purulent pleural effusion in both thoracic cavities, along with pleural thickening, and pustulation. Histopathology revealed hypertrophy of lung and thoracic wall serosa characterized by lymphatic infiltration, mesothelial hyperplasia, organizing fibrin deposition, and pseudomembrane formation. *P. canis* was isolated from organs, including pleural effusion. *P. canis* was detected in all 20 Ss, and *P. multocida* was detected in 16 Ss in them.

Conclusion The characteristic lesion of pasteurellosis (*P. canis* infection) in Ss in this case was chronic pleuritis. *Pasteurella* spp. are common resident in the oral cavity of Ss with high prevalence. From a zoonotic perspective, caution is warranted when rearing meerkats, particularly due to the risk of bites.

23: INVESTIGATION OF SYSTEMIC SKELETAL AND CARDIAC PATHOLOGY AND THE MECHANISM OF ANESTHESIA-INDUCED ARRHYTHMIA IN PORCINE DYSTROPHINOPATHY

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Background
Porcine dystrophinopathy has been reported in pigs with pale discoloration and fatty infiltration of skeletal muscle. However, the systemic distribution of lesions has not been fully documented. Of particular interest is its effect on cardiac function. We previously identified pigs that developed anesthesia-induced arrhythmia despite the absence of overt structural myocardial changes. This study aimed to evaluate skeletal and cardiac pathology and investigate the mechanism of arrhythmogenesis in affected pigs.
Materials and Methods
(i) Ten pigs judged as disposal of the whole body in meat inspection centers were examined gross pathology, histology (H&E), and dystrophin immunohistochemistry.
(ii) Arrhythmia induction was performed in Langendorff-perfused hearts from a carrier family (n = 10), with optical mapping conducted in four cases. Further analyses included dystrophin followed by Luxol fast blue staining, and N-cadherin/connexin 43 double immunohistochemistry.
Result
(i) The gracilis muscle showed the most prominent discoloration. Histology revealed

varying degrees of degeneration, fibrosis, and anisokaryosis in both skeletal and cardiac muscles, although myocardial lesions were milder. Dystrophin expression was reduced in both tissues.
(ii) Optical mapping during induced arrhythmia identified abnormal excitation foci at , which coincided with Luxol fast blue-positive, dystrophin-deficient regions. In adjacent areas, connexin 43 misalignment was observed, indicating focal gap junction disruption.
Conclusion
(i) Pigs with systemic skeletal muscle lesions due to dystrophinopathy also exhibit myocardial involvement.

(ii) Reduced dystrophin expression may impair electrical conduction through focal gap junction disruption, potentially leading to arrhythmogenesis under physiological stress conditions such as anesthesia.

24: AN UNUSUAL PRESENTATION OF FELINE PULMONARY LANGERHANS CELL HISTIOCYTOSIS IN A FOUR-YEAR-OLD CAT WITH CONCURRENT HEMOLYTIC ANEMIA

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A four-year old spayed female cat was referred to NC State's College of Veterinary Medicine for acute anorexia and regenerative anemia. Thoracic radiographs and abdominal ultrasound revealed disseminated coalescing nodules in the lungs and nodules infiltrating the kidneys. The lung and kidney nodules were aspirated and interpreted as highly suspicious for a carcinoma, and the animal was euthanized and submitted for necropsy. Gross evaluation of the lungs revealed coalescing, pale tan nodules disseminated throughout the lungs and similar nodules on the kidneys. Histologic evaluation of the lung revealed neoplastic histiocytes extensively invading and obliterating alveoli and terminal bronchioles. Similar neoplastic histiocytes were infiltrating the kidney and lymph nodes. Scattered throughout the liver, lymph node, and spleen were numerous, prominent hemophagocytic macrophages. Immunohistochemistry for IBA-1, E-cadherin, and pancytokeratin were applied to sections of lung, kidney, lymph node, liver, and spleen. Neoplastic histiocytes within the lung, kidney nodules, and lymph node were IBA-1 and E-cadherin positive and pancytokeratin negative, supporting Langerhans cell origin and feline pulmonary Langerhans cell histiocytosis (PLCH) in this patient. Several clusters of histiocytes in the spleen and lymph node were E-cadherin positive as well. Expression of E-cadherin by the hemophagocytic macrophages in the liver, spleen, and lymph node was unclear. This case represents an unusual presentation of feline PLCH due to the young age of this animal, primary presenting complaint of anemia, and concurrent IMHA. Electron microscopic evaluation of lung, liver, and lymph node is pending to further evaluate the histiocytes.

25: COMPARISON OF VISION TRANSFORMER AND RESNET MODELS FOR CLASSIFYING CANINE AND FELINE LYMPHOMA IN DIGITAL CYTOLOGY IMAGES

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Background: Cytologic evaluation of fine-needle aspirates is a routine, minimally invasive method for diagnosing lymphoma in dogs and cats. While deep learning offers promise for augmenting diagnostic accuracy, veterinary datasets are often limited, and systematic evaluations of modern architectures remain scarce.

Objective: To compare the performance of a Vision Transformer (ViT) and a convolutional neural network (ResNet-50) for binary classification of lymphoma versus non-lymphoma in canine and feline cytology images.

Methods: An in-house dataset of 306 digital cytology images (203 lymphoma, 103 non-lymphoma) was collected from canine and feline patients. Images were standardized and preprocessed identically for both models. The ViT-base patch16 (pretrained on ImageNet-21k) and ResNet-50 (pretrained on ImageNet) were fine-tuned on the cytology dataset using weighted cross-entropy loss and evaluated with 5-fold stratified cross-validation. Performance metrics included area under the receiver operating characteristic curve (AUC), accuracy, F1 score, and balanced accuracy. Model interpretability was demonstrated using attention maps (ViT) and Grad-CAM heatmaps (ResNet) and qualitatively assessed.

Results: ViT achieved mean AUC = 0.9239, accuracy = 0.8825, F1 score = 0.9145, and balanced accuracy = 0.8516. ResNet-50 achieved mean AUC = 0.9667, accuracy = 0.8659, F1 score = 0.8714, and balanced accuracy = 0.8673. Both models consistently highlighted diagnostically relevant lymphoid regions.

Conclusion: Both ViT and ResNet-50 demonstrated strong performance in classifying canine and feline cytology images, with complementary interpretability. These findings support the feasibility of extending this approach to additional cytologic tumor types and training general-purpose, modern deep learning classifiers to potentially aid or automate veterinary cytology workflows.

26: PERICARDIAL MESOTHELIOMA WITH LYMPH NODE METASTASIS IN A DOG

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A 7-year-old spayed female Cocker Spaniel presented to emergency service for suspected congestive heart failure. Clinical signs included tachycardia, weak femoral pulses, and muffled heart sounds. Thoracic radiographs revealed cardiomegaly; point-of-care ultrasound demonstrated significant pericardial and mild pleural effusion with evidence of cardiac tamponade; 270mL of presumptively hemorrhagic fluid was removed. On referral to cardiology service, echocardiography identified a mass in the right auricle and reaccumulation of pericardial effusion. A tissue hypoechoic relative to

the myocardium was noted adherent to the epicardium and pericardial surface. Given the guarded prognosis, humane euthanasia was elected. The heart and regional lymph nodes were submitted for evaluation. Grossly, the pericardium was thickened, pale tan – this appearance extended over the right heart; lymph nodes were markedly enlarged. Cytologic imprints revealed malignant neoplastic cells of undetermined origin. Immunocytochemistry with dual marker positivity (cytokeratin, vimentin) confirmed mesothelioma; cells were also seen in the lymph node imprints. Histologically, a moderately cellular neoplasm composed of polygonal epithelioid cells arranged in papillary and micropapillary projections lined the visceral pericardium and infiltrated the myocardium. Neoplastic cells were immunopositive for vimentin and cytokeratin, and were also identified in the diaphragmatic lymph node, indicating local metastasis. Mesotheliomas are rare neoplasms in dogs, typically arising from the pleura, peritoneum, or pericardium. Timely and accurate diagnosis remains challenging due to overlapping clinical and imaging features. Although metastasis is uncommon, it can occur, most often to regional lymph nodes. This case highlights the importance of including mesothelioma in the differentials of cardiac masses in dogs.

27: DIAGNOSTIC MICRORNA PANELS FOR CANINE VISCERAL HEMANGIOSARCOMA: TISSUE VALIDATION, MODEL SELECTION, AND BLOOD AND EFFUSION TESTING

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Background: Canine visceral hemangiosarcoma (HSA) is a highly aggressive vascular tumor with late-stage diagnosis limiting therapeutic success. Reliable and minimally invasive early diagnostic biomarkers are urgently needed.

Objectives: To validate diagnostic microRNA (miRNA) signatures in cardiac and splenic HSA tissues, identify optimal diagnostic panels, and assess their performance in blood/effusion samples.

Methods: Small RNA sequencing was performed on formalin-fixed, paraffin-embedded cardiac (n=6), splenic HSA tissues (n=18), and control counterparts (n=6 for each group). Forty candidate miRNAs were selected based on differential expression, fold change, and pathway relevance. Validation using customized qPCR panels confirmed 12 up- and one downregulated miRNA in cardiac, and five up- and eight downregulated in splenic HSA. To refine diagnostic potential, we applied model selection using Akaike Information Criterion (AIC). MiRNAs from top-performing models and validated miRNAs

were quantified using digital PCR in matched serum, whole blood, and abdominal and pericardial effusion samples from dogs with suspected HSA (n=8) and cancer-free (n=4).

Results: In serum, miRs- 493, 494, 376b, and 221 were upregulated in HSA-suspected dogs ($p<0.05$). Whole blood showed downregulation of miRs- 7a, 188, 221, 503, 34a, 362, 146a, and 150 ($p<0.05$). Expression patterns were consistent between whole blood and effusion samples, indicating both fluids are suitable for detection. Panel performance testing in liquid samples is pending.

Conclusion: This study identifies and validates tissue-specific and circulating miRNAs as diagnostic biomarkers for canine HSA. These miRNA panels may support the development of non-invasive tools to aid early and accurate diagnosis of this aggressive cancer.

28: DERMAL SPINDLE CELL PROLIFERATION WITH INTRALESIONAL MESOMYCETOZOEAN SPORES IN TWO CAPTIVE ADULT COHO SALMON (ONCORHYNCHUS KISUTCH)

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Formalin-fixed specimens from two adult Coho salmon from a public aquarium were submitted following the development of progressive skin lesions. One fish exhibited an ulcerated, soft, 16 x 10 x 8 cm mass over the mid-dorsum, with radiographs indicating soft tissue opacity. The other fish had multifocal small cutaneous nodules with scale loss and discoloration over the lateral body wall. In corresponding histologic sections from both fish, the dermis and underlying skeletal muscle were expanded by an unencapsulated, moderately to densely cellular, and infiltrative mass composed of spindle cells. The cellular infiltrate was arranged in interwoven streams, bundles, and whorls supported by variably collagenous to myxomatous stroma. Individual cells were fusiform with eosinophilic cytoplasm and round to elongate nuclei with coarsely stippled chromatin. Anisocytosis and anisokaryosis were moderate. There were ten mitotic figures in ten 400X high-power fields. Numerous 2–4 µm diameter, round, uninucleate spores were scattered throughout the spindle cell population. Spores stained positively with Grocott's methenamine silver and periodic acid–Schiff stains. In one salmon, similar spindle cell proliferation surrounded the kidney and there was granulomatous inflammation with abundant similar spores within the spleen, kidney, liver, heart, and gill. The morphologic features and staining characteristics of the spores were consistent with Mesomycetozoea, with *Dermocystidium* sp. or *Ichthyophonus* sp. considered most likely. Spindle cell proliferation closely resembled soft tissue sarcoma in these cases, a rarely described neoplasm in Coho salmon. The concurrent presence of Mesomycetozoea and a proliferative spindle cell response in salmonids has not been described previously.

29: ORAL HEMANGIOSARCOMA IN A CAT: A RARE PRESENTATION WITH DIAGNOSTIC AND COMPARATIVE SIGNIFICANCE

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A 10-year-old neutered male British Longhair cat presented with an ulcerated, infiltrative oral mass involving the right mandible. The lesion was locally destructive and bled easily on manipulation. Histopathologic examination revealed a densely cellular, unencapsulated spindle cell neoplasm forming irregular, blood-filled vascular channels. The neoplastic cells exhibited moderate anisocytosis, anisokaryosis, and a mitotic index exceeding 3 per high-power field. Nuclear protrusion into vascular lumina and the absence of well-formed vascular architecture complicated morphologic diagnosis. Immunohistochemical staining revealed strong cytoplasmic labeling for CD34 and Factor VIII-related antigen, confirming endothelial origin and supporting a diagnosis of hemangiosarcoma (HSA).
Oral HSA is extremely rare in cats and presents significant diagnostic challenges due to its atypical morphology and location. This case emphasizes the importance of combining histopathology with immunohistochemistry to establish a definitive diagnosis. Beyond its diagnostic interest, feline oral HSA offers translational relevance as a potential spontaneous model for human angiosarcoma. Insights into conserved tumor pathways—such as VEGF signaling, hypoxia-inducible factors (e.g., HIF-1α), and immune checkpoint dysregulation—suggest therapeutic targets that may benefit both species. Incorporating AI-assisted morphometrics and molecular profiling may improve diagnostic precision in ambiguous vascular tumors and support future case-based comparative studies. Finally, the rarity and histologic resemblance to human angiosarcoma suggest a valuable comparative model for future case-based studies in veterinary and human pathology.

32: IDENTIFICATION OF RECURRENT MUTATIONS IN CANINE MAST CELL TUMORS USING PCR AMPLIFICATION OF CYTOLOGIC SPECIMENS

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Background: Canine cutaneous mast cell tumors (MCTs) are common and show variable behavior from indolent to aggressive. Prognosis depends on histologic grade and metastatic potential. Mutations in the c-KIT proto-oncogene, especially exon 11, are linked to higher Kiupel grade, poorer outcome, and altered therapeutic response. A minimally invasive method of mutation detection may improve prognostication without surgical biopsy.

Objective: To evaluate the feasibility of detecting c-KIT mutations in archived cytologic MCT specimens using PCR and sequencing, and to assess the relationship between mutation status and Kiupel histologic grade.

Methods: Cytologic samples (n = 45) from canine MCTs were analyzed, including 15 low-grade, 15 high-grade (Kiupel), and 15 subcutaneous tumors. DNA was extracted, PCR amplification of c-KIT exon 11 was performed, and products were sequenced.

Results: PCR amplification was successful in 15/15 low-grade, 15/15 high-grade, and 14/15 subcutaneous tumors. Bands consistent with wild type c-KIT exon 11 were observed in 97.8% of all samples. Recurrent mutations were detected in 4/15 high-grade, 0/15 low-grade, and 1/15 subcutaneous tumors. Mutation frequency was therefore highest in high-grade tumors (26.7%) compared with low-grade (0%) and subcutaneous (0.067%), consistent with their poorer prognosis.

Conclusion: Cytologic samples allow reliable detection of recurrent c-KIT mutations in canine MCTs. Because mutation presence correlates with Kiupel grade and poorer prognosis, cytology-based mutation testing may serve as a minimally invasive tool for early prognostic assessment and treatment planning, reducing reliance on surgical biopsy.

33: EMERGING STAPHYLOCOCCUS AUREUS DISEASES IN NORTH AMERICAN BEAVERS

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Historically, Staphylococcus aureus has been considered a commensal organism with the potential for opportunistic disease in veterinary species, most notably mastitis or pyoderma. Recently, severe, multisystemic S. aureus infection has been described in Eurasian beavers (Castor fiber) infected with strains carrying a novel phage-borne bicomponent leukocidin, Beaver Leukocidin (BVL, lukF/S-BV), related to Panton-Valentine leukocidin (PVL, lukF/S-PV). Affected animals developed necrosuppurative pneumonia, nephritis, and disseminated visceral abscesses. Two North American beavers (Castor canadensis) submitted for necropsy to the Oregon Veterinary Diagnostic Laboratory had similar lesions, including abscesses in the lung, kidney, spleen, and heart. Aerobic cultures from affected tissues yielded heavy growth of S. aureus. Gross exam, histopathologic findings, and culture results mimic those reported in BVL-positive Eurasian beavers. Whole genome sequencing of S. aureus from one of the North American beavers identified previously undescribed leukocidin genes distinct from PVL. The similarity in lesion distribution and disease severity suggests these genes may be functionally analogous to BVL and contribute to pathogenesis. These parallels support the possibility of convergent evolution of virulence determinants in distinct S. aureus lineages. Herein, we describe the first report of severe, multisystemic S. aureus disease in North American wildlife. The identification of novel leukocidins in association with nearly identical pathologic patterns across species underscores the value of comparative pathology in detecting emerging pathogens and assessing their potential impact on animal and human health.

34: SPATIAL TRANSCRIPTOMICS APPLICATION IN FELINE FFPE SAMPLES USING THE PLATFORM GEOMX AND THE CANINE CANCER ATLAS (NANOSTRING-BRUKER)

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Background: Spatial transcriptomics explores gene expression in tissue architecture context. This technique has not been applied to cat samples.

Objective: To apply the spatial transcriptomics method in feline formalin-fixed paraffin-embedded (FFPE) samples, not previously reported.

Materials and Methods: Twelve FFPE samples of cat jejunum and ileum, with previous diagnosis of low-grade T-cell lymphoma or lymphoplasmacytic inflammatory bowel disease (six of each entity, supported by clonality analysis), and three control ileum samples were gathered. Freshly made 5-µm cuts were placed on positively charged slides. T-cell and B-cell populations were identified by immunofluorescence using anti-CD3 and anti-CD20 antibodies. The samples were processed using the GeoMx Digital Spatial Profiler for transcriptomic analysis with the Canine Cancer Atlas, which comprises 2010 targeting 1962 genes. A total of 92 regions of interest (ROI) were collected, each one including at least 100 positive cells for each T and B-cell marker for a total of 164 subfragments (aka Area of Illumination, AOI).

Results: Gene homology analysis indicated that 1574 cat genes shared >85% sequence identity. Both T and B-cell populations were clearly identifiable, and ROIs were successfully collected and sequenced. Quality control metrics flagged 6/164 AOIs as “Low-Stitched” and 7/164 as “Low-Aligned”. Additionally, the limit of quantification metrics, which assess the detection rate of each probe in ≥10% of the segments, showed that 1028/2010 probes met the criteria for further analysis.

Conclusion: Cat samples can be analyzed using the Canine Cancer Atlas, which allows for a deeper understanding of many feline oncologic diseases.

35: MAMMARY CARCINOMA AND PRESUMPTIVE PULMONARY METASTASIS WITH HYPERTROPHIC OSTEOPATHY IN AN ADULT FEMALE RACCOON (PROCYON LOTOR)

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An adult, female intact North American raccoon (*Procyon lotor*) was collected by wildlife officials and transferred to rehabilitation facility, where it was diagnosed with dehydration, emaciation, and limb stiffness. Radiographs identified a mass within the right caudal lung lobe and changes within all four limbs consistent with hypertrophic osteopathy. The raccoon was euthanized due to poor prognosis. On gross necropsy, multiple subcutaneous nodules are noted along the mammary chain and approximately 90-95% of the right caudal lung lobe is disrupted by a 9 x 5 x 5 cm, firm mass which is pale tan to dark red on cut section. On histologic evaluation, the mammary tissue is multifocally disrupted by a multinodular, encapsulated, well-demarcated, multicystic mass composed of epithelial cells arranged in sheets or tubules surrounded by swirling myoepithelial cells, consistent with a tubulopapillary mammary carcinoma. The pulmonary parenchyma is obliterated by a partially encapsulated, variably well-demarcated, highly cellular mass composed sheets of neoplastic cells representative of

the neoplastic population described in the mammary gland. The mammary tumor cells strongly immunolabeled for cytokeratin, while the pulmonary population displayed rare weak cytokeratin immunoreactivity and instead labeled strongly for vimentin. Neither population displayed immunoreactivity to CD204, IBA1, synaptophysin, chromogranin A, and TTF1, and non-neoplastic and neoplastic round cells did not label with CD18 IHC (not validated in raccoons). These findings are most suggestive of pulmonary metastasis of mammary carcinoma with epithelial to mesenchymal transition and secondary hypertrophic osteopathy.

36: THYMOMA-ASSOCIATED EXFOLIATIVE DERMATITIS IN AN AGED CAPTIVE BLACK BEAR (URSUS AMERICANUS)

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A 21-year-old, neutered male black bear (*Ursus americanus*) presented with progressive alopecia and pruritus unresponsive to weekly ivermectin therapy for suspected mange. Clinical deterioration included lethargy, inappetence, and labored respiration, which was followed by euthanasia and post-mortem examination. Necropsy revealed a cranial mediastinal mass measuring 30 cm x 17 cm x 9 cm compressing the thoracic structures and displacing the heart and lungs caudally. There was abundant scaling and alopecia diffusely affecting the skin. Additionally, there were multifocal lung mineralizations and peripheral and mediastinal lymphadenomegaly. Histopathology of the mediastinal mass identified a neoplasm most consistent with a Type B3 thymoma. Pustular and ulcerative dermatitis and epidermatitis with acantholytic cells, epidermal hyperplasia, dermal fibrosis, and orthokeratotic hyperkeratosis were identified within the skin. Additional histological findings included lymphoplasmacytic myocarditis and multifocal pyogranulomatous myositis. The constellation of dermatologic and muscular lesions is supportive of paraneoplastic syndromes that have been described in domestic species with thymomas, including cats, goats, and rabbits. This case represents a rare documentation of thymoma-associated exfoliative dermatitis and systemic paraneoplastic manifestations in a captive black bear. To our knowledge, this is the first report of its kind in *Ursus americanus*, expanding the comparative pathology of thymic neoplasms and their associated paraneoplastic syndromes in exotic species.

37: INDIRECT METHOD FOR REFERENCE INTERVAL DETERMINATION TO ASSESS BREED-SPECIFIC CHANGES - A PILOT STUDY

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Background: The use of novel statistical algorithms to indirectly estimate reference intervals (RI) from real-world veterinary patient databases has recently gained interest. However, their ability to detect breed-specific differences has not yet been fully investigated.

Objectives: To evaluate whether breed-specific indirect RI can accurately identify expected differences in hemoglobin (Hb) and white blood cell count (WBC) in adult Greyhounds.

Materials and Methods: Laboratory data were extracted from a mixed-breed patient database from IDEXX UK over a 20-month period. Indirect Greyhound-specific RI with confidence intervals (CI) were calculated for Hb and WBC using the RefineR algorithm, in dogs aged 1–8 years. These were compared to breed-specific direct RI established from a population of healthy Greyhound blood donors of the same age group in the same laboratory.

Results: For both Hb and WBC, indirect and direct greyhound-specific RI demonstrated similar lower and upper limits, with overlapping CIs, and were inside clinical equivalence limits, except for the upper Hb limit. Both RI sets differed substantially from non-greyhound RIs, with Hb higher and WBC lower in Greyhounds, matching clinical expectations and previous literature.

Conclusion: The indirect method successfully identified expected breed differences in Hb and WBC Greyhound RI, in agreement with the direct method – except Hb upper limit -and published data. This pilot study highlights the potential of indirect RI models as a practical, low-cost tool for determining breed-specific RI. Further research should explore their application across additional tests and the clinical value of breed-based RI stratification.

38: HIGH-PATH AVIAN INFLUENZA CAUSES CENTRAL NERVOUS SYSTEM NECROSIS IN NEW ENGLAND WILD BIRDS

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Since its North American incursion in 2021, highly pathogenic avian influenza (HPAI) has caused unprecedented morbidity and mortality in wildlife with an alarming degree of cross-species transmission. Unlike most avian influenza viruses, which primarily affect the gastrointestinal tract of waterfowl, this virus demonstrates increased neurotropism. Here we conducted opportunistic necropsies on wildlife with confirmed HPAI admitted to rehabilitation clinics or found deceased. A total of 42 necropsies were performed representing 17 avian species across 8 taxonomic orders. An animal's HPAI status was confirmed by RT-PCR, and sequencing when available. Of the necropsies with sequencing data (26), 25 were lineage C2.1 (H5N1) and 1 was lineage A6 (H5N5). Most animals that were observed or evaluated while alive exhibited neurologic clinical signs including ataxia, tremors, and nystagmus. Additionally, most birds were in decent body condition and of the animals with serology, 68% were influenza positive. Gross lesions were frequently observed in the brain, spleen, pancreas, and gastrointestinal tract.

Histopathological evaluation revealed extensive necrosis within the central system that corresponded to viral immunopositivity and presence of viral RNA by situ hybridization supporting the hypothesis that neurologic clinical signs stem from enhanced viral replication in the central nervous system. This study provides novel, multi-species documentation of severe, multi-organ pathology in naturally infected wildlife and underscores the urgent need for integrated wildlife disease surveillance, pathologic characterization, and genomic analysis to better understand host range, transmission dynamics, and zoonotic potential of emerging HPAI viruses.

39: 4',6-DIAMIDINO-2-PHENYLINDOLE IS A NOVEL STAINING METHOD FOR AMYLOID STAINING THAT BINDS TO BETA-SHEET STRUCTURE

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Background

Amyloidosis is caused by the extracellular deposition of amyloid fibrils with a β-sheet structure. 4',6-Diamidino-2-Phenylindole (DAPI) stain AA amyloid of bovine was found by chance. A recent study reported that DAPI stains amyloid. DAPI staining is simpler than Congo Red (CR) and Thioflavine T (ThT) staining; however, its staining mechanism remains unclear. Thus, this study aimed to investigate the mechanism and specificity of DAPI staining for amyloid and its utility.

Materials and Methods

The staining properties of DAPI and ThT for amyloid were compared based on the similarity of their chemical structures. To investigate the mechanism of DAPI staining for amyloid, formic acid-treated amyloid specimens were used to examine the mechanism through which structural changes influence DAPI binding to amyloid fibrils. DAPI staining was also evaluated in four specimens with different types of amyloid. The sensitivities of CR and DAPI staining were compared using amyloid specimens of different thicknesses.

Result

DAPI and ThT exhibited comparable chemical structures and staining characteristics. The amyloid found in formic acid-treated specimens did not stain with DAPI, indicating that DAPI recognizes the β-sheet structure. Conversely, DAPI staining was positive in specimens deposited with AA, Aβ, AL, and AIAPP. In thin specimens, DAPI was more sensitive compared with CR in staining amyloid.

Conclusion

DAPI staining can stain various types of animal amyloid. It recognizes the β-sheet structure of amyloid fibrils. Moreover, it is a simple and sensitive method for detecting amyloid deposition.

40: PATHOLOGICAL CHARACTERIZATION OF LETHAL AND NON-LETHAL LASSA VIRUS INFECTION IN A RHESUS MACAQUE MODEL

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Background: Lassa virus (LASV) hemorrhagic fever is an endemic, rodent-borne disease in many West African countries, causing up to 300,000 cases and 5,000 deaths annually. LASV infection is asymptomatic or mild in 80% of the population; however, hospitalized patients can have significant morbidity and a case fatality rate of up to 60%. Survivors commonly develop debilitating chronic sequelae including auditory, neurological, and ophthalmic manifestations, and alopecia.

Methods: Twenty-four rhesus macaques were subcutaneously inoculated with LASV on Study Day (DY) 1, with endpoints being death, moribund euthanasia, or terminal euthanasia (DY 29), followed by full necropsy, histopathological evaluation, and LASV immunohistochemistry on a subset of tissues.

Results: Animals clustered into three distinct lesion profiles (LPs). LP-1 (11/24 animals): early death or moribundity (DY 13-22) with multiorgan inflammation and necrosis, including interstitial pneumonia with pleural effusion and hepatocellular necrosis with intracytoplasmic viral inclusion bodies (ICIBs) in macrophages; additionally, there was multiorgan vasculitis restricted to capillaries, venules, and/or veins, often featuring fibrinoid necrosis. LP-2 (5/24 animals): later moribundity or survival to terminal euthanasia (DY 29) with vasculitis and vasculopathy involving the arterial system, often with robust inflammation; additionally, there was periarterial interstitial nephritis, glomerulitis, pancreatitis, and granulomatous mural folliculitis and necrosis associated with ICIBs in keratinocytes. LP-3 (8/24 animals): survival to terminal euthanasia (DY 29-30) with minor lesions.

Conclusion: LASV infection is associated with distinct microscopic lesion profiles, differing between survival outcomes. A novel finding of this study is the LASV-mediated mural folliculitis, which is potentially linked to alopecia reported in LASV survivors.

41: VALIDATION STUDY OF SMARTPATH MUSE TECHNOLOGY (SMARTPATH) FOR TISSUE-TO-DIGITAL IMAGING IN VETERINARY PATHOLOGY: A

CONCORDANCE STUDY COMPARING SLIDE-FREE VIRTUAL H&E WITH TRADITIONAL WHOLE SLIDE IMAGING

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Objective: This study evaluated diagnostic accuracy of SmartPath MUSE Technology™ slide-free imaging (SPI), generating virtual hematoxylin and eosin (H&E) images, compared to conventional H&E whole slide imaging (WSI) for veterinary surgical pathology.

Methods: One hundred deidentified companion animal biopsies (canine, feline, equine) were bisected to create mirror-image samples. One half underwent SPI, and the other was processed for standard H&E and scanned for WSI. Five board certified veterinary pathologists independently reviewed 1,000 total reads (500 SPI, 500 WSI) using a two-week washout period between modalities. Patient signalment per original test request form was provided. Diagnoses were categorized as concordant, minor discordance, or major discordance relative to a consensus reference. We assessed case and reviewer-level performance with sensitivity analyses.

Results: Using 495 cases per modality, one case eliminated due to poor SPI image quality, overall concordance was higher with WSI (95.6%, 95% CI: 93.4%–97.8%) compared to SPI (88.9%, 95% CI: 85.5%–92.0%), based on 95% bootstrap confidence intervals. With one outlier reader removed, the overall concordance for SPI improved to 91.9% (95% CI: 88.9%–94.6%), narrowing the gap with WSI (95.7%, 95% CI: 94.0%–97.8%) and reducing the between-modality difference to -3.8% (95% CI: -7.6%, -0.8%), suggesting that SPI performance was notably strengthened.

Conclusions: These findings support SmartPath Muse Technology™ slide-free imaging as a potential primary diagnostic modality in veterinary surgical pathology following appropriate validation and training. The technology's rapid, non-destructive workflow offers cost-effective alternative to conventional histology, enabling point-of-care applications, remote pathology review, and future integration with AI-assisted diagnostics.