



Late Breaker Poster

1: ISCHEMIC MYELOMALACIA SECONDARY TO MULTIFOCAL INTRAVASCULAR FIBROCARILAGINOUS EMBOLI IN THE SPINAL CORD OF A PONY MARE

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A 15-year-old pony mare presented to MSU-CVM's equine emergency service in lateral recumbency. She was up to date on vaccinations and housed in pasture with 3 full sized horses who are clinically normal. Upon presentation, she was unable to rise and began having focal seizures. Euthanasia was elected due to poor prognosis. On necropsy, she was in good body condition with minimal autolysis. There were no obvious gross lesions to explain her clinical signs. Rabies direct fluorescent antibody and meningeal aerobic bacterial culture were both negative. However, histopathologic evaluation of cervical spinal cord revealed severe, multifocal myelomalacia and hemorrhage. In multiple cervical cord segments, the grey matter horn (most often the ventral horn) and neighboring white matter were expanded by clear spaces, hemorrhage, and homogenous eosinophilic debris. Within affected gray matter segments, neurons were commonly angular and hypereosinophilic with pyknotic or absent nuclei and multifocally interspersed with swollen eosinophilic axons and small fibrin aggregates. Within the most affected section, a large cavitated tract was observed extending down a lateral funiculus. Within and surrounding this section, lateral and ventral funiculi contained dilated and often empty myelin sheaths. Multiple blood vessels within and marginating affected cord segments were multifocally occluded by intraluminal clear to amphophilic myxomatous matrix that strongly stained with Alcian blue (fibrocartilagenous emboli). Fibrocartilagenous embolic myelopathy occurs when embolization of nucleus pulposus fragments within spinal cord vasculature causing thrombosis and subsequent ischemic necrosis. Fibrocartilagenous emboli (FCE) are extremely uncommon in horses and only sporadically reported, typically in adult full-size breeds.

2: EX VIVO SCREENING PREDICTIVE TESTS FOR CANINE ONCOLOGY PATIENTS REPRESENT A PROMISING AVENUE FOR IMPROVING CANCER TREATMENT OUTCOMES IN DOGS

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It is estimated that one in four dogs will develop cancer throughout life, and those over 10 years of age have a 50% chance. The therapy of choice for cancer patients is usually surgery, which can be associated with chemo and/or radiotherapy. However, the standard treatment may not always be sufficient to control the disease in animal with higher clinical staging or irresectable tumors. Individualized treatment is an option that has been increasingly explored in human medicine, but not yet adopted in veterinary oncology. To establish personalized therapy, we used the *ex vivo* screening technique.

The aim of this study was to standardize the *ex vivo* screening test for the most used chemotherapy drugs, with the future perspective of translational medicine for cancer patients. We collected tumors from three dogs: one with pulmonary adenocarcinoma, one with a malignant mesenchymal tumor (both diagnosed by FNAC and immunohistochemistry) and the third one had an oral melanoma, with metastasis to RLN (diagnosis made by excisional biopsy). The samples were cultured in Matrigel, and cell viability (MTT) was tested for different chemotherapeutic agents. For the first dog carboplatin was the most effective, for the second dog sorafenib showed the best response and for the third, a combination of carboplatin and cyclophosphamide was chosen. All animals showed reduction in primary tumors and associated symptoms. This technique holds promise as an additional tool for predictive panels, facilitating personalized therapy for veterinary oncology patients.

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3: ACQUIRED RESISTANCE TO IXODES SCAPULARIS IN MUS MUSCULUS

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Ticks are obligate hematophagous parasites and pathogen vectors responsible for morbidity and mortality worldwide. Persistence and spread of tick-borne pathogens depend upon arthropod-host interactions, including immune responses to the tick itself. Non-native hosts can develop an immune response to ticks with repeated infestations, a phenomenon termed acquired rick resistance (ATR), which leads to rejection and/or death of the tick. In contrast, native tick-host pairings do not elicit ATR. Much of the research characterizing ATR has utilized guinea pigs (*Cavia porcellus*), which mount robust ATR in response to *Ixodes scapularis*. However, guinea pigs are distantly related to natural hosts and lack of species-specific regents hinders advancement in the field. *Mus musculus* reportedly lacks ATR with repeated *I. scapularis* infestations. However, our findings suggest that presence of ATR varies between strains. We monitored the feeding success of *I. scapularis* on C57BL/6 mice and histologically examined host bite sites and whole ticks. Repeated infestations correlated with decreased tick feeding success, and mice exhibited increasingly severe inflammation at tick bite sites characterized by mononuclear cells and polymorphonuclear cells with morphology indicative of eosinophils. Numbers of host leukocytes within midguts of replete larvae positively correlated with number of previous infestations experienced by the host. Host leukocytes were rarely observed in ticks fed on naïve hosts. Our findings contradict the established paradigm that *M. musculus* lacks ATR with repeated *I. scapularis* feedings. Identification of ATR in C57BL/6 mice provides new avenues for mechanistic investigation of ATR and will help facilitate tick-borne pathogen research and vaccine development.

4: CAN WE USE OUR PATIENT DATABASE FOR ESTIMATING REFERENCE INTERVALS? CHALLENGES AND OPPORTUNITIES OF THE NEW “INDIRECT METHODS”

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Background. Population-based reference intervals (RIs) are fundamental for appropriate result interpretation. Current recommendations for their determination specify that a direct a priori approach, including at least 120 carefully selected subjects, is preferable. This is not always feasible due to logistical and cost constraints. Indirect approaches using retrospective “big data” are well known since the 1960s but failed to provide accurate RIs. Recently, several sophisticated algorithms for the indirect estimation of RIs have been proposed in human medicine and described as comparable with direct methods in many cases.

Objectives. Preliminary evaluation of the utility and feasibility of novel indirect methods for determining RIs in the veterinary clinical laboratory.

Material and Methods. Four biochemical measurands with different indices of individuality (Albumin, ALP, Chloride, Creatinine) were chosen. Unselected patient results (n=100000) from a clinical laboratory were used to calculate RIs by applying three novel statistical algorithms: truncated minimum chi-square, tidyKosmic, and RefineR. The reference limits obtained were then compared to those currently in use in the laboratory, determined through a direct approach.

Results. The three novel algorithms provided almost identical reference limits. The reference limits were very similar to those directly established and generally inside their 95% confidence intervals. One algorithm was challenged with artificial pathological values up to 50% of the population and consistently returned identical reference limits.

Conclusion. Novel statistical tools for indirect estimation of RIs appear as a promising opportunity with good potential for sex/age/breed stratification and they may represent a cost-effective option when direct approaches are not feasible.

5: LESION PREVALENCE IN TOXICOLOGIC PATHOLOGY: FINDINGS AND USES OF LESION FREQUENCY DATA WITH ADJUSTABLE SEARCH PARAMETERS

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Introduction: Information on the prevalence of lesions in control animals is used for historical control databases and, along with the pathologist's own experience and data from the scientific literature, supports microscopic data evaluation. Similar data that also includes xenobiotic-treated animals may be used in additional strategic and research initiatives (i.e., developing decision support artificial intelligence algorithms, training to new staff, and implementation of virtual control groups).

Objective: Develop and apply a lesion prevalence algorithm.

Methods: We utilized SAP BusinessObjects software to query Provantis, our global laboratory information management (LIMS), and report lesion frequency based on several user selected metadata fields. Metadata parameters included: species, strain, duration of study, study group (control, treated), organ, animal age, and others. Diagnostic synonyms, such as “hyperplasia” and “increased cellularity” in each cell type were manually summed.

Results: Lesion prevalence was compiled based on the specified search parameters and qualified using 20 focused searches and verification of 500+ individual animal findings. Pathologist variability in diagnostic terms and the evolution of standardized terminology necessitated limited manual computations. Herein, we report the most common findings for four species in several common target organs.

Conclusions: This data provides insight into the overall prevalence of lesions. While it is impossible to provide and share all potential search permutations due to the complexity and scale of the datasets, this tool will allow CRL to provide datasets for highly specific use cases for our internal and external stakeholders.

6: INVESTIGATION OF NEONATAL HOOKWORM INFECTION ON IMMUNE VARIATION IN SOUTH AMERICAN FUR SEAL PUPS

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The immune system is highly variable between individuals, which results in variability in disease susceptibility and severity. Parasites have immunomodulatory effects that can influence susceptibility to or severity of concurrent infectious diseases. Our research aims to determine if early intestinal hookworm infections influence the immune phenotype of South American fur seals (SAFS).

SAFS are reproductively synchronized, and the majority are infected with intestinal hookworms (*Uncinaria sp.*) during their first days of life. Hookworms cause tissue damage and anemia in most species, but in eared seals (otariids) can be particularly aggressive, resulting in high mortality.

Fecal, blood and tissue samples were collected from 32 SAFS pups from birth and opportunistically recaptured over time. Using Luminex Bead-Based Assay, serum cytokine levels were assessed over time, and comparisons were made against hookworm fecal egg counts, hematological and biochemical parameters. Preliminary findings revealed a bimodal pattern in serum IFN γ expression: 10 pups had IFN γ levels that remained high after patent infection, and 13 pups had IFN γ levels that decreased over time. Pups without hookworm infections were used as control (n=9). These IFN γ patterns were positively correlated with IL-10 levels but not with fecal egg count, body mass, leukocyte, or biochemical parameters. This preliminary evidence suggests that pups with higher IFN γ levels at the time of hookworm infection (first 2 weeks of life) had decreasing IFN γ levels later in the season and were less likely to present with respiratory or gastrointestinal illness.

7: NECROHEMORRHAGIC PNEUMONIA IN A DOG CAUSED BY EXTRAINTESTINAL PATHOGENIC ESCHERICHIA COLI

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Acute necrotizing and hemorrhagic pneumonia caused by extraintestinal pathogenic *Escherichia coli* expressing cytotoxic necrotizing factors (CNF) 1 and 2 is a rare fatal disease of dogs. A 1-year-old female Labrador retriever presented to the Texas A&M Emergency & Critical Care Service following an acute onset of inappetence, dyspnea, hemoptysis, and fever. Radiographs revealed suspected bronchopneumonia and pneumothorax. The dog was euthanized due to a poor prognosis, and a necropsy was performed. Grossly, a hemorrhagic thoracic effusion was present, and all lung lobes were diffusely dark red and firm. Histologic examination of the lung revealed severe suppurative bronchopneumonia with necrosis, hemorrhage, and pleuritis with bacterial bacilli within macrophages and neutrophils. Hemolytic *Escherichia coli* was cultured from the lung, and toxin typing detected CNF 1 toxin. The intracellular location of *E. coli* was demonstrated with both immunohistochemistry and fluorescent in situ hybridization (FISH). Although extraintestinal pathogenic *E. coli* has been reported as a cause of pneumonia in dogs previously, this is the first report in which the bacterium has been demonstrated with immunohistochemistry and FISH.

8: MASSIVE PROLIFERATION OF GRANULATION TISSUE IN THE PARIETAL PERITONEUM OF A LABORATORY BEAGLE PREVIOUSLY IMPLANTED WITH A CARDIOVASCULAR TELEMETRY DEVICE.

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A 33-month-old male laboratory beagle was euthanized in part due to poor signal quality from a cardiovascular telemetry device that had been surgically implanted two years earlier. At necropsy, the parietal surface of the ventral peritoneum was covered in numerous irregular raised pink to red nodules ranging up to approximately 1 cm in diameter. This finding correlated histologically with multiple (occasionally confluent) pedunculated nodules composed of a variable-density collagenous stroma with numerous stromal cells arranged perpendicular to immature blood vessels (granulation tissue). This tissue was lined by a single layer of well-differentiated mesothelium. On immunohistochemistry, the mesothelial cells were positive for both vimentin and pancytokeratin, whereas the stromal cells were vimentin-positive but pancytokeratin-negative. Based on these immunohistochemistry results, we excluded the possibility of mesothelioma. The morphology of this finding was consistent with a florid nonneoplastic proliferation of granulation tissue rather than a neoplasm.

Granulation tissue may form in body cavities that are inflamed or implanted with foreign bodies. The granulation tissue proliferation in this dog did not occur at the site of the telemetry unit, electrode leads, or arterial catheter. However, parts of the device may

have migrated through the peritoneal cavity and could previously have been in contact with the affected portion of the body wall. Less likely, the finding could have developed at the site of the ventral midline incision through which the telemetry device was surgically implanted. This case illustrates the development of a potential reaction to intraperitoneally implanted telemetry devices.

9: EARLY MORTALITY ASSOCIATED WITH THE CARDIAC PHENOTYPE IN A NOVEL *FBN1*^{C1041G/+} *LOXL1*^{-/-} DOUBLE-MUTANT MOUSE MODEL OF IMPAIRED ELASTOGENESIS

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Background: Mouse models with fibrillin-1 (*Fbn1*) mutations are essential to the study of Marfan syndrome, which carries a high risk of aortic aneurysm and dissection. Deficiencies in lysyl oxidases such as *LOXL1* cause impaired elastogenesis and are also associated with aortic dilation and aneurysm. In this study, a novel mouse model was developed to examine ocular features as risk factors for glaucoma by crossing *Fbn1*^{C1041G/+} mice with *Loxl1*^{-/-} mice, creating double mutant (dbm) mice with both fibrillinopathy and impaired elastogenesis. Increased incidences of early mortality in these dbm mice at approximately 7-16 weeks old prompted further investigation in determining the cardiac phenotype.

Methods: Six wild type (wt) and 7 dbm mice were necropsied and the heart, pulmonary artery, and aorta were collected and fixed en bloc prior to imaging and histology processing. Hearts were examined grossly and histologically to evaluate cardiac chamber enlargement, aortic dimensions and ventricular wall thickness. Sections of the aortic root were stained with Movat histochemical stain to examine elastic fiber morphology.

Results and Conclusion: In all cases, gross examination showed cardiomegaly with severe bilateral atrial and aortic dilations in dbm compared with age-matched wt mice. Movat staining of the dbm aorta showed elastic fiber disorganization and fragmentation. While impaired elastogenesis has been reported in *Fbn1*- and *Loxl1*-mutant mice, the cardiac phenotype differs in the dbm mice. Further work and analysis are needed to investigate the underlying pathogenesis as well as other systemic manifestations associated with the mutations.

10: CANINE HIBERNOMAS: 91 CASES (2019-2023)

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Background: Japanese pathologists have periodically encountered hibernomas and noticed that the most instances occur in the muscles of chihuahuas. Canine hibernomas are still poorly understood.

Objective: We assessed the cytological, pathological, clinical, and epidemiological characteristics and tumor behavior of canine hibernomas.

Methods: A total of 91 hibernoma cases were examined. The biopsy submission forms were used to gather clinical data, and when available, follow-up data was also included. Seventeen cases were subjected to a panel of immunohistochemical stains.

Result: Chihuahuas (63.7%) were overrepresented. Hibernomas frequently develop in or near muscles: 33 cases of intramuscular or adhesion to the muscles of the legs, torso, and head; 12 cases of cutaneous trunci muscles; and 10 cases of tongue muscles. Four cases were ocular regions. Eight dogs had multiple hibernomas. Cytology revealed loosely cohesive polyhedral cells with a variable number of clear vacuoles and magenta-colored granules. Histologically, the masses were composed of sheets of polyhedral cells with abundant eosinophilic foamy cytoplasm. The majority were well demarcated, but 10 cases were infiltrative, and 5 cases had high mitotic counts. Immunohistochemistry, 16/17 expressed uncoupling protein 1 (UCP-1), 15/17 expressed desmin, and 12/17 expressed myogenin. Recurrence was noted in 5 cases, with a 10.6% recurrence rate. There were three metastatic instances. In one case, pulmonary metastasis led to a tumor-related death.

Conclusions: Chihuahuas in Japan are at increased risk of hibernomas, and canine hibernomas tend to occur in skeletal muscles. Even though the majority of canine hibernomas are curable following excision, some exhibit malignant behavior.

11: A DATABASE SURVEY OF EQUINE TUMORS AND IDENTIFICATION OF INDIVIDUAL RISK FACTORS FOR TUMOR DEVELOPMENT

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Background

Survey data of equine tumors are sporadic and specific demographic risk variables are poorly described. The incidence of different tumors changes over time and varies between countries making extrapolation from other dataset challenging.

Objectives

This study aimed to describe the most frequently diagnosed equine tumors from samples submitted to the UQ Veterinary Laboratory Services from 1980 to 2021 and identify any risk factor associated with the development of specific tumor types.

Methods

Records of all neoplastic equine submission were retrieved from the local database, categorised, coded according to a modified Vet-ICD-O-canine-1 coding system and analysed using a descriptive observational approach. Age was further analysed by one-way ANOVA with post-hoc Tukey-Kramer test. A multinomial logistic regression for breed, sex and age of horses with sarcoid and squamous cell carcinoma (SCC) was also performed.

Results

Of the 587 cases, the most frequently diagnosed tumors were sarcoid, squamous cell carcinoma (SCC), lymphoma, melanoma, papilloma. Most common sites were soft tissues, skin, eye and adnexa, as well as genital system. Out of 229 malignant cases, 12.2% were associated with metastases. Age was a significant discriminating factor between sarcoid and either lymphoma, SCC or melanoma. Geldings and Warmbloods were at increased risk of sarcoid, while stallions, Quarter Horses and Arabians had an increased risk of SCC.

Conclusions

By coding and classifying equine cancer data, specific risk factors for common cancer types were identified. This can allow for future targeted research into specific risk factors and a baseline to follow cancer trends over time.

12: DIGITAL KARYOMETRY OF CANINE URINARY BLADDER UROTHELIUM DEMONSTRATES THAT NUCLEAR SIZE AND SHAPE DEFINE MALIGNANCY

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Criteria for cancer diagnosis are often based on abnormalities of the cell nuclei. However, the definition of nuclear atypia is often based on descriptive factors, which can complicate the assessment in diagnostics. In the age of artificial intelligence (AI), measurements of cell nuclear parameters can be performed rapidly in an objective, quantitative, and reproducible way. The aim of this study was to measure the dimensions of cell nuclei of benign (normal or hyperplastic) (n=20) and malignant (urothelial carcinoma) (n>200) canine bladder urothelium by use of a commercially available AI-histology software (Visiopharm, Horsholm, Denmark). H&E stained histology glass slides were scanned, urothelial regions were defined on these digitized slides, and nuclei were detected automatically with AI software. From each nucleus, the following parameters were measured by the software: area, form factor (level of roundness), perimeter, axis (minor, major), and diameter (largest, lesser). Preliminary results demonstrate that the nuclear area is statistically significantly larger in tumor tissue when compared to hyperplastic and normal tissue (malignant 42.2 μm^2 , hyperplastic 33.4 μm^2 , normal 26.9 μm^2 ; $p < 0.001$). Likewise, the perimeter is significantly larger in malignant tissue compared to benign tissue (malignant 24.7 μm

versus benign 21.3 μm ; $p < 0.001$) and the form factor is reduced in malignant compared to benign nuclei ($p < 0.001$).

Our preliminary data show that nuclear size and shape are significantly different between benign and malignant urinary bladder urothelium. Moreover, such measurements can be easily and quickly performed with commercially available AI histology software to define malignancy of canine urothelium in a diagnostic setting.

13: GLOSSITIS AND PERIODONTITIS IN C57BL/6 MICE: A HAIRY AFFAIR

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We report glossitis and periodontitis in C57BL/6NJ mice during routine histopathology evaluation as part of a high throughput phenotyping screen. Focal and multifocal glossitis was mostly localized to the base of the tongue at the level of the lingual salivary glands. The mild to moderate neutrophil- and macrophage-rich inflammation with scattered lymphocytes and occasional mast cells was present in the epithelium, lamina propria, and muscle bundle layers. Periodontitis was characterized by a similar mixed inflammatory infiltrate within the gingival sulci of the maxillary molars frequently resulting in gingival ulceration, periodontal alveolar bone resorption, new alveolar bone formation, and infrequently complete loss of the molar. Areas of glossitis and periodontitis typically surrounded hair shaft fragments.

We conducted this retrospective case series study to determine the frequency and concurrence of glossitis and periodontitis and any causal association with the presence of hair shaft fragments. We identified glossitis and periodontitis in 149 (28.8%) and 130 (25.1%) of the 517 mice, respectively. The two pathologies were concurrent in 92 (17.8%) of the 517 mice; this association was statistically significant ($p < 0.001$). Intralesional hair shaft fragments were identified in 138 (92.6%) and 124 (95.4%) of glossitis and periodontitis lesions and was significantly associated with both pathologies ($p < 0.001$), confirming penetration of tongue and gingival epithelium as the etiology. Potential implication for animal health and phenotyping is discussed.

14: MAMMARY CARCINOMA AND MALIGNANT MYOEPITHELIOMA IN A GERIATRIC CHINCHILLA

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Background: A mass from the right inguinal mammary gland of a 13-year-old, intact female chinchilla.

Objective: To characterize a case of mammary carcinoma and malignant myoeptithelioma in a chinchilla.

Results: Macroscopically, the mammary gland was expanded by a tan, firm, ulcerated, multilobulated mass. Microscopically, the neoplasm was biphasic and composed of distinct epithelial and spindle cell populations with moderate mixture between the cell types. Neoplastic epithelioid cells were arranged in islands and cords, sometimes exhibiting squamous differentiation and forming large cystic spaces containing keratin, on a dense fibrovascular stroma, whereas neoplastic spindle cells were arranged in interweaving fascicles on a fine fibrovascular stroma. Within both populations, neoplastic cells had indistinct cell borders, vacuolated cytoplasm, and an oval vesicular nucleus with up to two nucleoli. Anisocytosis and anisokaryosis were marked, with 49 and 35 mitotic figures in the epithelial and spindle cell populations, respectively, in 10 high power (400x) fields. Within the spindle cell population, neoplastic cells had variable expression of myoepithelial cell markers, including diffuse immunoreactivity for vimentin, frequent immunoreactivity for cytokeratin-14, and occasional immunoreactivity for smooth muscle actin and pan-cytokeratin. Within the epithelioid population, neoplastic cells had diffuse immunoreactivity for pan-cytokeratin and additional immunoreactivity for cytokeratin-14 only within the basal cell layer. The neoplastic cells were therefore confirmed to belong to two distinct populations, with the spindle cells representing pleomorphic interstitial myoepithelial cells.

Conclusions: To our knowledge, this is the first report of mammary carcinoma and malignant myoepithelioma in a chinchilla.

15: DETECTION OF HIGHLY PATHOGENIC AVIAN INFLUENZA H5N1 IN THE BRAIN OF DOMESTIC CATS AND WILD MAMMALS IN OREGON

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Background

In November 2022, three outdoor domestic cats were housed near a poultry flock with a confirmed HPAI outbreak. Within a week, one cat went missing and two developed morbidity resulting in death or euthanasia. In May 2023, two skunks were located next to a raptor carcass (confirmed HPAI positive); one was dead, the other moribund. A fox kit was found in labored breathing and expired shortly thereafter.

Objective

To provide a detailed description of the pathology associated with HPAI H5N1 in 2 domestic cats, 2 striped skunks, and a red fox in the USA.

Methods

An Avian Influenza Matrix PCR, Avian Influenza Virus Subtype H5 and clade 2.3.4.4 rtPCR were performed on brain tissue, which was confirmed by NVSL. Routine necropsy and histopathology were performed on all animals.

Results

Brains from all animals were rabies negative. Cat lungs and brains of wild animals tested positive for HPAI H5N1 EA/AM reassortment 2.3.4.4 clade by OVDL and NVSL testing.

Grossly, both cats exhibited severe pneumonia. No specific gross lesions were observed in the skunks and fox.

All animals had necrotizing endomyocarditis/myocardial necrosis. Other lesions included lymphocytic meningoencephalitis (cat 2, skunks, fox); necrotizing bronchopneumonia (cats, skunks); necrotizing pancreatitis (cat 1; skunks); and adrenalitis, hepatic or renal necrosis (single animals each).

Conclusion

Avian influenza is now established as an endemic pathogen worldwide, and now poses a risk to mammalian species in close contact with infected birds. This case constitutes the first confirmed case of HPAI H5N1 in domestic cats in the USA.

16: COMMON TUMORS AND TUMOR-LIKE LESIONS OF PET RODENTS AND MMTV DETECTION IN MAMMARY TUMORS OF PET RODENTS IN TAIWAN

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Compared to many references described neoplasms in lab rodents, fewer studies focused on the neoplasms in pet rodents. Of note, mouse mammary tumor virus (MMTV) might be associated with tumors of mammary gland in rodents. This study collected 77 tumors from 70 pet rodents, including hamsters (48/70), guinea pigs (16/70), unknown species (3/70), rats (2/70), and gerbil (1/70). Overall, the most common affected system was skin (62/77), followed by reproductive system (7/77) and urinary system (3/77). The most common tumor was adenocarcinoma of mammary gland tumor (15/77), followed by fibrosarcoma (10/77) and lipoma (5/77). 50 tumors were collected from hamsters, and the most common tumor is mammary tumor (13/50), followed by fibrosarcoma (9/50), mast cell tumor (4/50) and squamous cell carcinoma (4/50). The collected subtypes of mammary tumor included tubular carcinoma (n=5), tubular adenoma (n=4), carcinoma and malignant myoepithelioma (n=1), simple tubular carcinoma (n=1), adenosquamous carcinoma (n=1), and tubulopapillary adenoma (n=1). 20 tumors were collected from guinea pigs, and the most common tumor is lipoma (6/20), followed by adenocarcinoma of mammary gland (4/20). In guinea pigs, the subtypes of mammary gland tumor were tubular carcinoma (n=2), tubular and solid carcinoma (n=1), and tubulopapillary carcinoma (n=1). Of 20 cases of mammary tumors, MMTV was not detectable via nested polymerase chain reaction. This study described the common tumors in pet rodents and revealed no evidence of MMTV infection in mammary gland tumors in pet rodents in Taiwan.

17: DEVELOPMENT OF AN AI-ASSISTED CYTOLOGIC TUMOR GRADING MODEL FOR CANINE CUTANEOUS MAST CELL TUMOR

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Background: Mast cell tumor (MCT) is the most common cutaneous neoplasm of dogs. A published cytologic grading scheme shows relatively good correlation with histopathologic grade but is time consuming and prone to inter-observer variability. Artificial intelligence (AI) technology could potentially increase efficiency and accuracy of cytologic grading.

Objective: Develop an AI-assisted model for cytologic grading of canine cutaneous MCTs.

Methods: Digitally scanned cytologic preparations from a variety of canine cutaneous MCTs were utilized for computer vision training. Whole slide images were broken into tiles to facilitate training. Mast cells, nuclei, multinucleated cells, and mitotic figures were annotated by board certified pathologists. Object detection techniques included Mask Region Convolutional Neural Network and Deformable Detection Transformer. The trained model was evaluated using mean average precision, Jaccard index, and precision, and improved based on validation outcomes and pathologist review of identified and classified objects. Logistic regression was used to identify correlations between findings and histopathologic grade. Once fully optimized, the model will be employed on a larger testing set with known histopathology grades.

Results: Initial evaluation of the model shows good to excellent precision (80% or greater) for identification of mast cells, nuclei, multinucleated cells, and mitotic figures. The number of mitotic figures and multinucleated cells per mast cell show positive correlation with high grade.

Conclusion: Our AI model can successfully identify features commonly used to cytologically grade canine cutaneous MCTs. This AI-assisted approach presents a promising advancement in accurate and efficient cytologic tumor grading.

18: A REVIEW OF 77 CASES OF IDIOPATHIC NECROTIZING ENTERITIS (INE) IN BEEF CALVES

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Background: Idiopathic necrotizing enteritis (INE) was first described during an outbreak of enteric disease in 3-month-old beef calves in Scotland in 1994. It has since been diagnosed throughout the UK causing diarrhea, pyrexia, and tachypnea. Reported

gross findings include gastrointestinal ulceration, pneumonia and pale kidneys. The underlying etiology remains unknown and current literature is limited to individual case reports.

Objective: To complete a retrospective analysis of INE cases and identify epidemiological features, clinical signs and pathology associated with INE.

Methods: Pathological reports and archived glass slides of 77 cases of INE diagnosed between 2012-2022 were reviewed. Data were extracted and collated based on previously published characteristics and on novel features detected.

Results: INE was predominantly diagnosed in spring-born beef calves (98%) with an average age of 11 weeks and average 7-day duration of clinical signs. Clinical signs included diarrhea (61%), dehydration (55%), tachypnea (32%), pale mucous membranes (22%), pyrexia (15%), and dermatitis/ alopecia (10%). Gross pathological findings included ulceration of laryngeal tonsils, esophagus, stomachs and intestines (especially small intestine, 85%). Other pathological changes included pneumonia (69%), renal changes (60%) and fibrinous pleuritis (32%).

Conclusions: Epidemiological characteristics, clinical signs and gross findings in calves diagnosed with INE were predominately consistent with historic case reports. Gastrointestinal ulceration was frequent. Ulceration of other less expected organs, pneumonia, fibrinous pleuritis, and renal changes were more common than anticipated, and not all cases presented with diarrhea. Re-evaluation of the archived slides for common histopathological features is ongoing to establish the microscopic features associated with INE.

19: DEVELOPMENT OF A NOVEL DECISION SUPPORT TOOL FOR DETECTION OF CARDIAC ABNORMALITIES IN THE NON-HUMAN PRIMATE USING PATHOLYTIX AI

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Introduction: Utilizing decision support tools focused on abnormality detection can increase the efficiency and consistency of the toxicologic pathologist's workflow.

Objective: Use supervised training to develop a decision support tool that detects and highlights abnormalities in the non-human primate heart.

Methods: Normal and abnormal tissue classes were annotated on H&E-stained whole slide images (WSI) of non-human primate (NHP) hearts at 40x magnification. Annotations were ingested by a deep machine-learning convolutional neural network provided by Patholytix AI and a classifier was developed that identified inflammation/infiltrate, cardiomyocyte degeneration/necrosis, and adipose atrophy/degeneration/necrosis. Model performance was assessed using F1-score metrics, confusion matrices and visual inspection by an expert pathologist. A qualified

rat heart classifier was applied on the NHP hearts for comparison and to discern if transfer learning could aid in accelerating improvement of the NHP model.

Results: The NHP classifier identified lesions with sensitivities ranging between 0.84 to 0.98. Precision for the inflammation/infiltrate class was lower than the other classes, which was due to involvement with heterogeneous lesions (encompassing several classes). The performance of a qualified rat heart classifier in the non-human primate heart suggested that the classifier built in the rat may be a good starting-point for the NHP heart classifier.

Conclusions: Utilization of this tool by toxicologic pathologists may increase the efficiency and consistency of workflow while reviewing digital slides. Continuing work will focus on creating a more robust model, adding more lesion classes and annotations, and further adaptation to detect cardiac abnormalities across other organs and species.

20: EFFECTS OF ORAL AMBROXOL ON NEUROPATHOLOGICAL MEASURES IN THE CANINE MODEL OF MUCOPOLYSACCHARIDOSIS TYPE IIIB

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Mucopolysaccharidoses (MPSs) are lysosomal storage diseases caused by enzyme deficiencies leading to glycosaminoglycan (GAG) accumulation. Some MPS types, including MPS I, II, III, and VII, affect the nervous system and cause accumulation of heparan sulfate (HS) and gangliosides. MPS IIIB, characterized by alpha-N-acetyl glucosaminidase (NAGLU) deficiency, primarily results in HS accumulation and lacks approved treatments. Gene therapy is desirable to address NAGLU deficiency effectively. Additionally, an adjunct medication with widespread distribution, enhancing enzyme activity and slowing substrate accumulation, may yield better outcomes.

Ambroxol therapy benefits patients with neuronopathic Gaucher disease by increasing enzyme activity, reducing substrate buildup, and improving neurological parameters. We hypothesized that ambroxol therapy would ameliorate neuropathological changes, decrease lysosome volume, and reduce GAG accumulation in MPS IIIB. Three groups of animals were analyzed: unaffected (n of 2), untreated MPS IIIB affected (n of 2), and ambroxol-treated MPS IIIB affected (n of 1) canines. The treated animal received oral ambroxol hydrochloride, 30 mg/kg/day divided q12 hours, from 2 to 16 weeks of age. LIMP2 immunolabeling was used to assess GAG accumulation indirectly by analyzing lysosomal volume, while GFAP and Iba1 were used to assess gliosis. Quantitative analysis revealed reduced LIMP2, GFAP, and Iba1 immunolabeling in the treated animal's brain and spinal cord compared to untreated controls. HS levels were similar to untreated controls. Although preliminary, these findings suggest a potential positive effect of ambroxol on MPS IIIB disease.

21: POST MORTEM FINDINGS OF LARGE FELIDS KEPT UNDER HUMAN CARE

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The study provides an overview of post mortem findings observed in 33 large felids kept under human care in a variety of different facilities (zoological gardens, rescue center, circus) located in North-Eastern Italy. The collection included 10 tigers, 8 leopards, 7 lions, 4 caracals, 1 lynx, 1 bobcat, 1 serval, 1 cheetah (age-range: 1 day-20 years).

Inflammatory changes were frequently encountered in the gastrointestinal system (24/33 animals, 73%) mainly in the gut (16/24). Ulcerative gastritis was detected in 7 animals (3 young harbored gastric foreign bodies). Renal lesions (23/33 animals, 70%), especially in adult/old animals, were predominantly interstitial, followed by tubular and glomerular. Pulmonary lesions, frequently encountered (19/33 animals, 57%) were mainly inflammatory, followed by alveolar edema and anthracosis. Liver lesions (17/33 animals, 51%) included vacuolar hepatopathy, hepatitis and hepatic cysts. Degenerative and inflammatory joint diseases were detected in 6 adult or old animals (18%), while 6 felids exhibited dilated cardiomyopathy. Three young animals displayed cardiopulmonary filariasis with microfilaremia. Tumors were found in 2 leopards: 1 thyroid adenoma and 1 mesothelioma. One cheetah presented systemic amyloidosis and one caracal pup showed severe hydrocephalus associated with severe diffuse purulent meningitis and encephalomyelitis.

The statistical analysis (Fisher-Yates and Chi-squared tests) suggested: a prevalence ($p < 0.05$) of age-related lesions in tigers and caracals; a prevalence of infective/parasitic lesions in puppies; males showed more traumatic lesions than females.

Summarizing, lesions of the gastrointestinal tract, pneumonia and renal inflammatory and degenerative changes represent the most frequent findings in our collection of captive large felids.

22: TISSUE DISTRIBUTION OF HIGHLY PATHOGENIC AVIAN INFLUENZA IN COLORADO WILDCATS

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Background: Highly pathogenic avian influenza (HPAI) has decimated domestic poultry and been detected in multiple mammalian species in the United States. In Colorado, HPAI has been detected in several free-ranging wildlife species including bobcats (*Lynx rufus*) and mountain lions (*Puma concolor*), raising concerns both for wildlife health and human exposure to harvested carcasses. Tissue distribution of HPAI in wild felids is not well understood and is of interest to help direct HPAI surveillance in these species.

Objective: We assessed the lesions and tissue distribution of HPAI in 4 mountain lions (*Puma concolor*) and 2 bobcats (*Lynx rufus*) from Colorado using histopathology and immunohistochemistry (IHC)

Methods: IHC performed using a goat polyclonal antibody anti-Influenza A (Abcam ab20841). Our method was validated and confirmation with outside diagnostic validated IHC (University of Georgia Veterinary Diagnostic Laboratory).

Results: Histologic lesions were identified in the brain (6/6), lung (4/6), liver (4/6), gastrointestinal tract (2/6), adrenals (2/6), kidney (1/6), skeletal muscle (0/6). The most common lesion was necrotizing meningoencephalitis (5/6 animals).

DISCUSSION: Histologic lesions associated with HPAI infection in free-ranging mountain lions and bobcats from Colorado most frequently occurred in the brain (100%), and commonly in the lung and liver (67%), with the most common lesions being necrosis followed by lymphoplasmacytic inflammation. Immunohistochemistry confirmed the presence of Influenza A in the cytoplasm and nuclei of inflammatory cells associated with necrotizing lesions.

23: IMPACT OF COMMONLY ENCOUNTERED MEDICAL CONDITIONS ON CELL-FREE DNA CONCENTRATION IN THE PLASMA OF DOGS WITHOUT CANCER

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BACKGROUND

The concentration of cell-free DNA (cfDNA) in plasma is elevated in dogs with cancer. Past studies suggest that conditions other than cancer may cause elevations in canine cfDNA concentration.

OBJECTIVE

This study evaluates whether the presence of concomitant conditions at the time of blood draw impacts plasma cfDNA concentration in dogs without cancer.

METHODS

Blood samples were subjected to cfDNA extraction and quantification using TapeStation (Agilent), then classified/aggregated by whether the patient had one or more comorbidity present at blood draw. Samples were identified with disease [n=312: top aggregates were allergy (single condition n=54; with other conditions n=98), musculoskeletal/lameness (n=46; n=94), cardiac disease (n=22; n=42), infection (n=14; n=40)] or without disease (n=593). Groups were compared using Wilcoxon Sign Rank Test, p-values <.005 were considered significant following Bonferroni Correction.

RESULTS

Samples from dogs with cardiac disease (as single condition) showed significantly lower cfDNA concentrations than dogs without disease ($p=.004$). When multiple conditions were present in the same patient, no significant increase in concentration was seen for infection ($p=.015$), and no significant decrease for cardiac and renal disease ($p=.017$ and $p=.011$, respectively).

CONCLUSIONS

This data suggests that patients with cardiac disease have decreased cfDNA concentrations compared to dogs without concomitant disease; further studies should explore biologic underpinnings. Though commonly encountered conditions of routine severity may not significantly impact cfDNA concentration, additional studies are needed to evaluate clinical information (severity, flare up, treatment). Multiple concurrent conditions may be more likely to impact cfDNA concentration, though significance was not reached in this study.

24: IS THIS A PENILE TERATOMA? WHAT'S YOUR MORPHOLOGIC AND PHILOSOPHIC RATIONALE?

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Background/Objective: A 3-week-old, male, C57BL/6J mouse had a small pink growth on his head on dorsal midline. The animal was humanely euthanized and submitted to the UC Davis Comparative Pathology Laboratory for post-mortem examination and histopathology of the growth.

Methods: The mass was formalin-fixed, paraffin-embedded, and stained with routine hematoxylin and eosin (HE) and pancytokeratin, CK14, CD31, and smooth muscle actin.

Results: Arising from the dermis is a well-demarcated, exophytic mass comprised of multiple tissue types and overlaid by keratinizing stratified squamous epithelium. This epithelium transitions into nonkeratinizing stratified squamous epithelium as it bilaterally (presumed circumferentially) invaginates, recapitulating prepuce. Medial to the epithelial invaginations, the surface mid-mass further invaginates and recapitulates the urethral groove as the nonkeratinizing stratified squamous epithelium merges into transitional epithelium, recapitulating urethra. Surrounding the central canal are blood sinuses, lined by anastomosing trabeculae of endothelium and muscle, reminiscent of corpus spongiosum. Immunophenotyping confirmed the embryologic ontogeny of tissues from ectoderm, mesoderm, and endoderm.

Conclusions: The mass on the head is consistent with penile tissue. The mouse was confirmed to have a normal penis present. The authors vigorously debated definitions and ontogenic considerations and came to the consensus this mass is a teratoma. The tissues are derived from multiple germ layers and are heterotopic, which is defined as normal tissue arising in an abnormal location. A hamartoma derives from one germ

layer. Choristoma was considered, but because most of the mouse urethra is derived from endoderm, a teratoma confers the most accurate diagnosis given the available facts.

25: CAUSES OF DEATH IN CAPTIVE SNAKES FROM PRIVATE ENCLOSURES IN THE NORTH-EAST OF ITALY

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Recently, the public interest on snakes is drastically increased, in parallel with the number of exotic animals housed as pets. A variety of snake species are kept under human care and, whereas these animals' pathology isn't fully investigated, the main causes of disease rely on infections, following by nutritional imbalances or deficiencies, while neoplasia are rarely reported.

This study aims to investigate, through a complete standardized post-mortem investigation, the causes of mortality in 13 captive boids and 12 pythons dead between 2019 and 2023 in zoological gardens and households.

12 snakes showed signs of severe inflammatory diseases; the target organs were lungs (6) and kidneys (3), liver (2) and skin (1). 15 snakes were affected by degenerative hepatic conditions (lipidosis, glycogenosis and cysts). 1 python showed multiple masses of a rectal carcinoma with hepatic metastases, while 1 boa presented a severe encephalitis. To date, 19 snakes were RT-PCR-tested for Reptarenavirus: 8 showed a multiorgan positivity, while 3 showed a single organ positivity. Histologically, the viral inclusion bodies related to inclusion body disease were clearly and diffusely visible in 5 of these animals. *C. serpentis* was detected both in feces and histologically within gastric epithelium of one boa, and oocysts referable to *Caryospora* spp. and *Coccidia* spp. were found in 4 animals, respectively.

In conclusion, Reptarenavirus remains an endemic disease in many snakes and needs continuous diagnostic attention, as well as infections, but the importance of management conditions should be underlined to owners to proper maintenance.

26: ECHINOCOCCUS MULTILOCULARIS IN A MUSKRAT (ONDATRA ZIBETHICUS) IN SOUTHERN ONTARIO, CANADA

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Echinococcus multilocularis is a tapeworm that can cause alveolar echinococcosis (AE), a public health concern and a fatal disease in human and mammals. Recently, Southern Ontario has been recognized as a risk area for *E. multilocularis*, and as of 2018, *E. multilocularis* has been designated as a reportable disease in Ontario. However, it is

unknown which wildlife species are affected by this parasite. Muskrats are important furbearer of North America and are harvested commonly by trappers with hunting dogs. Bodies of 159 muskrats were submitted to the Southern Ontario Ministry of Northern Development, Mines, Natural Resources, and Forestry (MNR) between 2022 and 2023 as part of ongoing muskrat health surveillance research. During postmortem examination, one muskrat had multiple hepatic and splenic cysts. *Echinococcus multilocularis* was confirmed through characteristic histopathological features of multiple hydatid cysts that contained metacestode protoscolices with calcareous corpuscles, and rostellar hooks, along with polymerase chain reaction (PCR). This case is the first report of *E. multilocularis* in muskrat in Southern Ontario, suggesting wide spread of *E. multilocularis* in Southern Ontario, with increased zoonotic risk towards fur harvesters. In addition, it highlights the importance of surveillance for early detection of zoonotic diseases in wildlife populations, and emphasizing the role of muskrats as sentinel species in new emerging zoonotic disease.

27: ATYPICAL INTRAVASCULAR LYMPHOMA IN A DOG

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Intravascular lymphoma (IVL) is challenging to diagnose antemortem, as large neoplastic lymphocytes proliferate within the lumina of small blood vessels in the absence of overt leukemia, accumulation within lymph nodes, or formation of discrete masses. This report is an investigation of IVL in a 12-year-old female spayed, mixed breed canine that presented with jejunal perforation. A protracted clinical history included two jejunal resections and anastomoses. Histopathologic evaluation of excised tissue indicated focal necrosis and mild lymphoplasmacytic jejunitis. Cytologic evaluation of peritoneal effusion fluid obtained after a third exploratory laparotomy yielded a diagnosis of lymphoma in addition to suppurative inflammation. Neoplastic lymphocytes were ~40 µm in diameter, with rare azurophilic cytoplasmic granules, and amoeboid nuclei. Flow cytometric evaluation of peritoneal fluid leukocytes showed a population of large CD4/CD45/MHCII⁺ lymphocytes suggestive of T, natural killer (NK), or NKT cells. A concurrent CBC indicated immune-mediated hemolytic anemia without presence of atypical lymphocytes. Due to clinical deterioration, the dog was euthanized. At post-mortem examination, there was marked peritonitis. Histologic evaluation revealed large neoplastic lymphocytes expanding the serosa and muscularis mucosa of the small and large intestine, near and within vessels of the liver, kidney, lung, brain, and various lymph nodes, but neither forming aggregates nor being present in the bone marrow. Immunohistochemical (IHC) analysis on ante-mortem jejunal and post-mortem tissues revealed the intravascular lymphocytes to be CD3⁺/granzyme-B⁺/CD79a⁺/PAX5⁺. These results illustrate the challenge of ante-mortem diagnosis of IVL. Whether IVL in this case constitutes a T, NK or NKT cell proliferation remains to be determined.

28: DEVELOPMENT OF ARTIFICIAL INTELLIGENCE-ASSISTED MACROPHAGE IDENTIFICATION IN CANINE DIFFUSE LARGE B CELL LYMPHOMA: A PILOT STUDY TO INVESTIGATE PROGNOSTIC IMPLICATIONS

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Background:

Canine diffuse large B cell lymphoma (DLBCL) is the most common neoplasm in dogs. Within the tumor microenvironment, tumor-associated macrophages play a pivotal role and have been associated with worse prognosis in humans. These macrophages can engage in the phagocytosis and scavenging of tumor cells, resulting in the characteristic “starry-sky” pattern by the presence of tingible body macrophages (TBM).

Objective:

This investigation aimed to apply artificial intelligence (AI) for histopathology image analysis and determine the degree of TBM infiltration into DLBCL for further correlation analysis of their impact outcomes.

Method:

The analysis included retrospective cases of canine DLBCL (present in lymph nodes or spleen), regardless of age, breed, or sex, submitted to the Virginia Tech Animal Laboratory Services (2013–2023). Of 924 cases, 43 were selected due to the availability of complete clinical history. Pictures of ten fields within the neoplastic population were taken in a randomized manner and used for annotation, training, and analysis using QuPath. The cases were further divided into two groups based on survival (<6 and ≥6 months).

Result:

The AI successfully counted TBM in 34 cases (79%), while 9 cases were identified as outliers (exceptionally high or low counts). No correlation was observed between the numbers of TBM and survival outcomes.

Conclusion:

The AI effectively identified TBM, although, in a few instances, excessive stroma and autolysis were confounding factors. Future studies will aim at AI refinement, confirmation of TBM with immunohistochemistry, and a larger data set to yield more precise results.

29: INHAND: INTERNATIONAL HARMONIZATION OF NOMENCLATURE AND DIAGNOSTIC CRITERIA FOR LESIONS - AN UPDATE – 2023

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The INHAND Proposal (International Harmonization of Nomenclature and Diagnostic Criteria for Lesions in Rats and Mice) has been operational since 2005. A Global Editorial Steering Committee (GESC) coordinates objectives of the project. Development of terminology for rodent organ systems or non-rodent species is the responsibility of Working Groups, with experts from North America, Europe, and Japan. All rodent organ systems have been published – Respiratory, Hepatobiliary, Urinary, Nervous Systems, Male Reproductive and Mammary, Zymbals, Clitoral and Preputial Glands and Hematolymphoid System in Toxicologic Pathology and the Integument and Soft Tissue, Female Reproductive System, Digestive System, Cardiovascular System, Skeletal System, Special Senses and Endocrine System in the Journal of Toxicologic Pathology as supplements and on a web site – www.goReni.org. Mini-pig and Dog have been published in Toxicologic Pathology in 2021 and Non-human primate and Rabbit have been published in the Journal of Toxicologic Pathology in 2021. Fish and Non-rodent ocular toxicity group are targeted to have a manuscript for review in 2023. INHAND guides offer terminology, diagnostic criteria, differential diagnoses, images, and guidelines for recording lesions in toxicity and carcinogenicity studies. INHAND GESC representatives work with representatives of FDA Center for Drug Evaluation and Research (CDER), Clinical Data Interchange Standards Consortium (CDISC), and National Cancer Institute (NCI) Enterprise Vocabulary Services (EVS) to incorporate INHAND terminology as preferred terminology for SEND (Standard for Exchange of Nonclinical Data) submissions to the FDA. Interest in INHAND nomenclature, based on input from industry and government scientists, is encouraging wide acceptance of this nomenclature.

30: GLUCOCORTICOID RECEPTOR AGONISTS AS A NEW THERAPEUTIC APPROACH FOR GASTROINTESTINAL STROMAL TUMOR IN DOGS, AN IN SILICO STUDY

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Gastrointestinal stromal tumors (GISTs) are rare gastrointestinal mesenchymal neoplasms that occur in humans, dogs, and other species. In dogs, they are more frequently found in the intestine, while in humans, they are more common in the stomach. These tumors are associated with mutations in *c-KIT* and *PDGFRA*. While targeted therapy with tyrosine kinase inhibitors can be effective, it doesn't work for all patients. To explore alternative treatment options, drug repositioning shows promise.

The aim of this study was to analyze the global gene expression profile of canine GISTs, a tumor with limited available information. We used seven samples of canine intestinal GISTs and five normal canine intestine. Global gene expression analysis was conducted using GeneChip® Canine Gene 1.0 ST Arrays and Transcriptome Analysis Console 4.0.2.15 software to analyze the data (ThermoFisher Scientific). The results

revealed 109 upregulated and 252 downregulated genes that are involved in pathways like cell migration and proliferation.

In a novel step, the identified genes were input into the LINCS L1000FWD platform. This analysis led to the identification of glucocorticoid receptor agonists as a potential group of drugs that could potentially reverse the gene signature associated with canine GISTs.

Overall, this study contributes to the advancement of new therapeutic approaches for GISTs and enhances our comprehension of this neoplasm in dogs. Given that dogs serve as a valuable natural model for comparative oncology studies, these findings hold significant promise for both veterinary and human medicine.

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31: IMPACT OF OPIOID ANALGESIA TREATMENT ON PANCREATITIS OUTCOMES IN AN EXPERIMENTAL MOUSE MODEL

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Background: Disease conditions that are painful in people are painful in animals. However, the alleviation of pain in animal models may impact the experimental outcome. Obesity worsens pancreatitis in humans. **Objective:** A pilot study was done on the effect of opioid analgesia on the initiation and progression of pancreatitis in obese mice. **Methods:** IL-12/18 pancreatitis was induced *ob/ob* mice, $n = 9$ male; (weight 57.5 ± 9.5 g; Body Fat 25.0 ± 7.0 g) and $n = 3$ female; (46.8 ± 11.6 g; Body Fat 21.9 ± 4.2 g), with six mice ($n = 5$ male; $n = 1$ female) being treated with Ethiq-XR (Buprenorphine), 3.25 mg/kg, subcutaneously, and six mice ($n = 4$ male; $n = 2$ female) not receiving analgesia. Mice were euthanized at 52-69 hr, depending on the endpoint. **Results:** Baseline carotid pulse distention (CPD) (mm), rectal temperature ($^{\circ}\text{C}$), serum lipase (U/L), and calcium (mg/dL) measurements were 405 ± 88 mm, 36.9 ± 0.8 $^{\circ}\text{C}$, 98.3 ± 87.9 U/L, and 9.6 ± 1.8 mg/dL, respectively. Mice receiving Ethiq-XR had the following outcomes at 48 hours: CPD; 246 ± 21 μm , temperature 32.9 ± 2.2 $^{\circ}\text{C}$, terminal lipase, $2,314 \pm 965$ U/L, and calcium of 8.2 ± 2.5 mg/dL. These were 138 ± 32 mm, 28.4 ± 1.8 $^{\circ}\text{C}$, $6,710 \pm 2,144$ U/L and 3.8 ± 0.8 mg/dL respectively in mice not receiving Ethiq-XR (P -value < 0.01 vs. Ethiq-XR group for all parameters). Buprenorphine thus impaired the increase in serum lipase, and reductions in CPD, rectal temperature and serum calcium, indicating interference with the induction and progression of pancreatitis. Grossly, Ethiq-XR caused a large reduction in abdominal fat necrosis. **Conclusions:** This data supports that buprenorphine interferes with induction and progression of IL12/18- pancreatitis in obese mice.

32: CUTANEOUS T- CELL RICH LARGE B- CELL LYMPHOMA WITH ASSOCIATED AMYLOIDOSIS IN A HORSE

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A 12-year-old Quarter Horse presented with a 3 cm in diameter mass on the left mandible. No peripheral lymphadenopathy, pain, or other clinical signs were observed. An excisional biopsy was performed and submitted for routine histopathologic examination. Microscopically, the mass was composed of neoplastic round cells mixed with abundant small lymphocytes, fewer macrophages, plasma cells, and occasional multinucleated giant cells. Mixed with these cells were multifocal to coalescing, anastomosing trabecular deposits of extracellular amorphous, homogenous, eosinophilic material (amyloid). Neoplastic cells were round or had bizarre nuclei and occasional mitotic figures (6-9/2.37mm²). Congo red staining and immunohistochemical (IHC) analysis with CD3, CD20, Pax5, Mum1, and Iba-1 were performed. IHC revealed strong and diffuse positivity of CD3 of the small lymphocytes. There was about 20% positivity of Mum-1, Pax5, and Iba-1 among these round cells. Pax5 labeling identified neoplastic cells as B-cells. Amorphous material stained with Congo red and showed apple-green birefringence under polarized light. PCR for antigen receptor rearrangements (PARR) was performed at Colorado State University. PARR identified clonally rearranged immunoglobulin light chain genes which was consistent with a B-cell neoplasm or plasmacytoma. A diagnosis of TCRLBCL with amyloid production was achieved. T- cell rich large B-cell lymphoma (TCRLBCL) is the most common subtype of cutaneous lymphoma in horses. Cutaneous amyloidosis occurs rarely in horses and was a differential, however, the IHC and PARR results are not consistent with amyloidosis. Here we report a TCRLBCL associated with amyloid which appears to be a novel finding in the horse.

33: COX-2 IMMUNOREACTIVITY, REGIONAL INFLAMMATION AND METASTASIS IN CANINE APOCRINE GLAND ADENOCARCINOMA OF THE ANAL SAC

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Background

COX-2 enzyme expression has previously been identified in canine apocrine gland anal sac adenocarcinoma (AGASACA). COX-2 has a variety of tumorigenic effects, including promotion of inflammation and metastasis.

Objective

To determine if COX-2 immunolabelling was positively associated with anal sacculitis, intratumoral inflammation, metastatic spread and overall survival in canine AGASAC.

Methods

The study population was 47 retrospective cases from a single referral center of canine AGASAC that had a histological diagnosis with full oncologic staging, treatment information and follow up. Cases underwent COX-2 immunohistochemistry alongside histological review of intra-tumour and anal sac inflammation. Correlations were defined

using Spearman's Rank analysis. COX-2 results were also introduced into a multivariate Cox regression backwards elimination model of prognostic histological parameters.

Results

COX-2 immunoreactivity was present in 64% (30/47) of the canine AGASAC. In this study population, COX-2 immunoreactivity, measured by a semi-quantitative composite score combining intensity and area, did not correlate with severity of intratumoral or anal gland inflammation, nor extent of metastatic spread (primary only, local lymph node, distant metastasis). In a multivariate model, COX-2 expression was not significantly associated with altered survival when considered alongside the previously identified prognostic factors of solid histological pattern, intratumoral necrosis and vascular invasion.

Conclusions

COX-2 immunoreactivity in canine AGASAC did not correlate with inter-tumoral inflammation, anal sac inflammation, extent of tumor metastasis, or overall survival in the study population.

34: AFGHAN HOUND NECROTIZING MYELOPATHY IN A TERRIER MIXED BREED DOG

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Background: Afghan hound necrotizing myelopathy, also known as Kooiker leukodystrophy and hereditary necrotizing myelopathy, is a devastating myelinolytic spinal cord disease that causes destruction of the white matter in spinal cord with progressive spinal degeneration. The disease is first reported in Afghan hounds and reported in miniature poodles, Kooiker dogs, and other crossbreeds. Disease pathogenesis is presumed to be metabolic or enzymatic defect involving myelin that is of autosomal recessive inheritance.

Case presentation: A 1-year-old, spayed female Terrier mixed breed dog was presented to the University of Illinois Emergency Service for progressive nonambulatory tetraparesis, tremors, and episodes of panting with hypersalivation. Magnetic resonance imaging revealed bilaterally symmetrical intramedullary lesions within the white matter at the ventral and dorsal aspects of the cervical spinal cord, suggesting leukodystrophy/leukomyelopathy. Humane euthanasia was elected due to the declining quality of life and uneventful recovery from anesthesia. Postmortem examination revealed extreme softening and liquefactive changes in all funiculi in the white matter of spinal cord (leukomyelomalacia) with evident myelin vacuolation, florid myelinolysis, and numerous gitter cells. The lesions were bilaterally symmetrical in ventral, lateral, and dorsal funiculi and most pronounced throughout the thoracic segment and, to a lesser extent, the cervical segment with sparing of the lumbar spinal cord.

Conclusion: The present findings of characteristic bilaterally symmetrical necrotizing leukomyelopathy and early age onset of disease are compatible with those described in Afghan hound necrotizing myelopathy.

35: IRIDAL DEFORMITY IN FREE-RANGING JUVENILE RED-TAILED HAWKS (BUTEO JAMAICENSIS)

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Vision is a critical element of survival for free-ranging raptors, and those with impaired or lost eyesight face significantly higher mortality rates. Ocular disease is amongst the most common findings in captured, free-ranging raptors that present for clinical care, of which, trauma is overrepresented. Between 2006-2016, eight juvenile red-tailed hawks that were admitted to The Raptor Center of the University of Minnesota underwent postmortem examination for ocular lesions. The irises of all affected birds were bilaterally misshapen in the form of a horizontally oblong appearance to the pupils. Histologically, all affected eyes exhibited ectropion uveae, had pre-iridal fibrovascular membranes, and had dispersed pigment within the trabecular meshwork. Additionally, in some cases there were a seemingly increased number of pigment cells in the iris stroma, retinal atrophy, fibroplasia of the trabecular meshwork, and/or fibrous connective tissue spanning the drainage angle. All cases presented during October and November, coinciding with peak fall migration along the Mississippi flyway. Given the apparent predilection in red-tailed hawks, the possibility of a congenital etiology is in question; however, because of the frequency of trauma, particularly in young birds, these lesions may represent the aftermath of head trauma with ocular injury.

36: AVIAN MYELOCYTOMATOSIS VIRUS AND SHAMONDA VIRUS IN COMMON RAVENS (CORVUS CORVAX) WITH POLYOSTOTIC OSTEOMAS

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Background: From 2014-2022, 16 free ranging common ravens (*Corvus corvax*) from Maine (n = 13), Massachusetts (n = 2) and New York (n = 1) were brought to various wildlife rehabilitation centers. All animals presented bright and alert but unable to fly. They developed limited mobility and onset of depression. During necropsy the common lesion was multifocal, irregular to spherical, mineral-density nodules in the periosteal and endosteal surfaces contiguous with cortical bone. Histopathology was consistent with osteomas or exostoses consisting of well-differentiated, anastomosing networks of

woven bone lined by a single layer of osteoblasts and osteocytes.

Objective: To identify an underlying pathogen using next generation sequencing.

Methods: Tissue samples (2 spleen, 1 heart, 5 liver, 5 bone, 2 lung, 5 kidney) from 5 affected individuals were utilized. Extracted RNA from each organ was sent for library prep, ribodepletion and RNA sequencing to the HudsonAlpha Genome Sequencing Center.

Results: Bone samples contained the highest number of phyla. Alpharetrovirus (Avian myelocytomatosis virus, or AMV) was present in multiple sample sites but abundant in bone tissue only. Shamonda orthobunyavirus was dominant in all samples except bone where AMV was prevalent.

Conclusions: This is the first known report of avian myelocytomatosis virus in the common raven. Shamonda orthobunyavirus is present in most birds in multiple tissues and is potentially unrelated to the bone lesions. Ongoing work includes assessing tissues from remaining cases for presence of the noted viruses, and spatial genomics to characterize the distribution of the identified sequences in lesions.

37: ATYPICAL PERIPHERAL ODONTOGENIC FIBROMA IN A DOG: A CASE REPORT AND DISCUSSION.

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A 9-year-old spayed female Golden Retriever dog was evaluated for an expansile gingival mass adjacent to the left maxillary canine. Debulking surgery was performed, and the biopsy was reported as sarcoma. Local recurrence was reported 10 days later. A second biopsy was performed approximately 4 months after recurrence and was interpreted as atypical peripheral odontogenic fibroma (POF) due to the presence of stellate cells in a periodontal-like matrix with hard matrix material, yet relatively increased cellularity, mildly increased pleomorphism, and mitotic activity. Computed tomography (CT) scan demonstrated a 1.8 cm x 3.6 cm x 4.5 cm osseous mass with nasopharyngeal extension. Due to concern of recurrence, hemimaxillectomy was elected. Histopathology of the rostral hemimaxilla was consistent with the prior biopsy diagnosis of an atypical POF. Perioperative clinical staging demonstrated no overt metastasis. In serial follow ups of 1 month, 2 months, 3 months, 8 months, and 13 months from the original diagnosis, no local recurrence nor metastases were appreciated. A retrospective review of 19 dogs with diagnoses of atypical POF had similar findings; local recurrence was appreciated in only 2 patients, and metastasis was not reported in any case (Median follow up time 16.5 months). Six dogs had died or were euthanized due to unrelated disease with a median survival time (MST) of 20 months. Atypical POF may be an important differential when considering mesenchymal proliferations in the oral cavity of dogs. Our data indicates that atypical POFs have similar clinical behavior and prognosis to typical POFs.

38: PRESUMPTIVE PELGER-HUËT ANOMALY IN A BROWN-THROATED SLOTH (*BRADYPUS VARIEGATUS*)

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Background: The Pelger-Huët anomaly (P-H) was initially described in humans during the early 20th century by two German researchers. It has since been identified in some species, such as dogs, cats, rabbits, and horses. In humans, it manifests as an autosomal dominant trait resulting from a mutation in the gene encoding the lamin B receptor, impairing nuclear segmentation and heterochromatin trafficking, especially in granulocytes. In this condition, a significant nuclear hyposegmentation and a condensed to coarse chromatin pattern can be observed. It is crucial to differentiate between inflammatory or myelodysplastic conditions and true P-H, as this anomaly typically does not affect leukocyte functions or cause clinical signs.

Objective: This study aims to describe the blood cells of a Brown-Throated Sloth (*Bradypus variegatus*) with abnormal morphology within leukocytes.

Methods: An EDTA blood sample was obtained from a healthy *B. variegatus* in 2022 and submitted for checkup analysis. Complete blood count (CBC) was performed using an automated veterinary hematology analyzer. Blood smears were stained with Diff-Quik™ and evaluated by a trained veterinary clinical pathologist under light microscopy.

Results: In terms of CBC findings, the examination of blood smears revealed hyposegmented neutrophils and eosinophils. These cells displayed oval to band-shaped nuclei with mature, condensed nuclear chromatin. There were no observable toxic alterations within the granulocytic lineage. All remaining CBC variables were unremarkable.

Conclusion: This is the first report describing a presumptive P-H anomaly in *B. variegatus*. Future molecular investigations should be conducted to identify specific gene mutations for this species.

39: EVALUATION OF A RAPID IMMUNOCHROMATOGRAPHIC ASSAY FOR THE DIAGNOSIS OF RABIES IN REGIONAL LABORATORIES OF COSTA RICA

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Rabies is a viral, lethal, and zoonotic disease with worldwide distribution, primarily transmitted through bites from infected dogs and bats. The Biosafety Laboratory (LSE-LANASEVE) of the Animal Health Service of Costa Rica (SENASA) serves as the

national reference laboratory for diagnosing rabies in humans and animals. Since regional laboratories lack the equipment for the direct fluorescent antibody test (FAT), we evaluated the rapid immunochromatographic diagnostic test (RIDT) from BioNote, employing FAT as a reference, to improve rabies diagnosis. [Methodology] We analyzed 193 brain tissue samples between 2014 and 2019. Out of these, 174 came from species that RIDT has been validated for: bovines (162), dogs (10), and raccoons (2). The rest were from unvalidated species, including horses (7), humans (1), and others. [Results] Among the 174 validated samples, 26 bovine samples were positive for both RIDT and FAT. Reviewing all 193 samples, 28 were positive and 165 negative using both methods. Two horse samples presented inconsistencies, being positive on FAT but negative on RIDT; these were subsequently verified as false negatives by RT-PCR. RIDT exhibited a sensitivity of 94% (CI95, 83.9-102.3), specificity of 100%, positive predictive value (PPV) of 100%, and negative predictive value (NPV) of 99% (CI95, 97.1-100.5). [Conclusions] RIDT has demonstrated reliability in quickly diagnosing rabies for validated species. We advise its application in SENASA's regional laboratories for those particular species. If there's uncertainty, samples should be sent to LSE-LANASEVE for FAT or RT-PCR confirmation.

40: ENHANCING HEPATOCELLULAR HYPERTROPHY DIAGNOSIS IN RATS: AI-DERIVED NUCLEAR DENSITY MAPS IMPROVE DIAGNOSTIC CONSISTENCY

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Background: Xenobiotic exposure often triggers hepatocellular hypertrophy in rats, which may be challenging to diagnose consistently. Computer-assisted diagnostics (CAD) can provide decision support and is an active area of research. Hepatocyte size can be estimated by AI nuclear segmentation and whole-slide level density maps. This study evaluated the sensitivity, specificity and the impact on the pathologist's diagnostic consistency of AI-derived nuclear density maps for hepatocellular hypertrophy in rats.

Methods: Liver slides from Sprague-Dawley rats in an archived xenobiotic 28-days toxicity study were digitized at 40x using a whole slide scanner (WSS). A generic nuclear segmentation algorithm was modified with tailored size, shape and color filters to enhance identification of hepatocyte nuclei over other. Detection thresholds for nuclear density were determined separately for male and female subjects, referencing untreated animals. Slides were assessed by an unblinded veterinary anatomic pathologist before and after a four-day washout period, with and without density maps. Sensitivity (Se), specificity (Sp), positive predictive value (PPV) and negative predictive value (NPV) for hypertrophy detection via whole-slide density maps were calculated. Grading repeatability, comparing aided and unaided scoring was evaluated using Cohen's weighted kappa.

Results: Se, Sp, PPV and NPV for hypertrophy detection using whole slide density maps were 0.83, 0.91, 0.71 and 0.96 respectively, compared to unaided

scoring. Unaided pathology scoring demonstrated a Cohen's weighted kappa of 0.64, while aided pathologist scoring reached 0.94.

Conclusion: AI-derived nuclear density maps enable detection of hepatocellular hypertrophy in rats on a low-power slide grid, enhancing scoring repeatability for hepatocyte hypertrophy.

41: EFFECTS OF ORAL AMBROXOL ON NEUROPATHOLOGICAL MEASURES IN THE CANINE MODEL OF MUCOPOLYSACCHARIDOSIS TYPE IIIB

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Mucopolysaccharidoses (MPSs) are lysosomal storage diseases caused by enzyme deficiencies leading to glycosaminoglycan (GAG) accumulation. Some MPS types, including MPS I, II, III, and VII, affect the nervous system and cause accumulation of heparan sulfate (HS) and gangliosides. MPS IIIB, characterized by alpha-N-acetyl glucosaminidase (NAGLU) deficiency, primarily results in HS accumulation and lacks approved treatments. Gene therapy is desirable to address NAGLU deficiency effectively. Additionally, an adjunct medication with widespread distribution, enhancing enzyme activity and slowing substrate accumulation, may yield better outcomes. Ambroxol therapy benefits patients with neuronopathic Gaucher disease by increasing enzyme activity, reducing substrate buildup, and improving neurological parameters. We hypothesized that ambroxol therapy would ameliorate neuropathological changes, decrease lysosome volume, and reduce GAG accumulation in MPS IIIB. Three groups of animals were analyzed: unaffected (n of 2), untreated MPS IIIB affected (n of 2), and ambroxol-treated MPS IIIB affected (n of 1) canines. The treated animal received oral ambroxol hydrochloride, 30 mg/kg/day divided q12 hours, from 2 to 16 weeks of age. LIMP2 immunolabeling was used to assess GAG accumulation indirectly by analyzing lysosomal volume, while GFAP and Iba1 were used to assess gliosis. Quantitative analysis revealed reduced LIMP2, GFAP, and Iba1 immunolabeling in the treated animal's brain and spinal cord compared to untreated controls. HS levels were similar to untreated controls. Although preliminary, these findings suggest a potential positive effect of ambroxol on MPS IIIB disease.

42: COMPARISON BETWEEN CYTOLOGIC RESULTS OF EAR SWAB SAMPLES COLLECTED FROM DOGS AND CATS STORED IN AMIES LIQUID UP TO 5 DAYS

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Effects of examination time delays on canine and feline ear cytologic results of swabs—qualitative (presence/absence) and semiquantitative (scale, none to 4+) evaluation for bacteria, yeast, ear mites, and epithelial cells—are relatively unknown. Objective was to determine whether ear swabs stored at room temperature for up to 5 days in sterile

tubes each with a sponge moistened with the preservative Amies liquid affected the cytologic diagnostic quality.

Three swabs were collected from each of 12 dog ears and 8 cat ears. All swab samples were immediately placed into sterile tubes containing a sponge moistened with Amies liquid and then randomly selected for microscopy on day zero (collection day; D0), day 3 (D3), and day 5 (D5). Laboratory technicians used standardized, semiquantitative scales to score samples for bacteria (cocci and rods), yeast, epithelial cells, and ear mites.

Among samples, differences in scaled scores over the 5-day storage period were minimal: cocci 15%, rods 10%, yeast 10%, and epithelial cells 5% of an increase of at least 1 scaling increment. No directional trends were observed over time. The largest score changes were observed for cocci but considered minimal with a precision of 85% among all time groups. Changes in scale between time groups for yeast and rods were low, with a precision of 90%. Samples had minor changes in epithelial cells with a precision of 95%. No mites were detected.

Amies sponge tubes may be acceptable for storing canine and feline ear swab samples for up to 5 days for diagnostic application.

43: PATHOLOGIC FINDINGS IN 11 STRIPED SKUNKS DIAGNOSED WITH HIGHLY PATHOGENIC AVIAN INFLUENZA FROM PRINCE EDWARD ISLAND, CANADA

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In the ongoing North American outbreak, H5 highly pathogenic avian influenza virus (HPAI) was first detected in late 2021 in Atlantic Canada, causing neurologic signs and high mortalities in wild and domestic birds. It has since been detected in a range of avian and mammalian species in the region and across North America. Between November 2022 and May 2023 in Prince Edward Island, Canada, 13 striped skunks presented for necropsy at the Canadian Wildlife Health Cooperative, Atlantic, pathology service and 11 were confirmed positive for either H5N1 (n=10) or H5N5 (n=1) HPAI. All were adults in good body condition. Skunks displayed neurologic clinical signs, such as seizures, tremors and circling, lethargy and weakness, or were found dead. Common gross lesions included pulmonary (n=10) and nasal turbinate (n=5) edema, interstitial pneumonia (n=10), multifocal pulmonary (n=4) and hepatic (n=3) necrosis, pulmonary hemorrhage (n=4) and meningeal congestion (n=3). Microscopic lesions included meningoencephalitis (n=10), gliosis (n=11), pulmonary edema and congestion (n=10), necrotizing hepatitis (n=9), cerebral and brainstem neuronal necrosis (n=8), and bronchiointerstitial pneumonia (n=8). These lesions are consistent with previously reported lesions in skunks infected with H5N1 HPAI though this cluster of cases includes the first description of H5N5 HPAI in striped skunks. The high proportion of HPAI-positive skunks submitted for necropsy from a small geographic area over winter

and spring suggests skunks were feeding on HPAI-positive birds at relatively high rates during this time.

44: DETECTION OF HEMOTROPIC MYCOPLASMAS (HEMOPLASMAS) IN INDIGENOUS INDIVIDUALS OF BRAZIL

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Hemotropic mycoplasmas (hemoplasmas) are small, pleomorphic, uncultivable bacteria that may cause anemia in several mammalian species, including humans. Indigenous populations may be exposed to hemoplasma infection due to their contact with animals and vectors, with no study to date. Accordingly, the present study has assessed hemoplasmas infection in Brazil's Indigenous populations after approval by the Ethics Committee in Human Health of the Brazilian Ministry of Health. Overall, ten indigenous communities from Southern Brazil were sampled, with 23/632 (3.6%) positive individuals to hemoplasmas by qPCR ($C_t \leq 34.4$). However, only one case was confirmed by sequencing of the 16S ribosomal RNA gene, which indicated *Mycoplasma haemocanis* or *M. haemofelis* infection, as both species have identical sequences of this region. To the author's knowledge, this is the first cross-sectional epidemiological approach to hemoplasma infection in Indigenous populations and report of hemoplasma infection in a person from Indigenous communities. Previous hemoplasma infections were also reported in immunocompromised individuals or people with contact with animals and travelers, including *Mycoplasma haemofelis*, *Mycoplasma suis*, *Mycoplasma haemohominis*, *Mycoplasma ovis*, and *Mycoplasma haematoparvum*. The hemoplasma infection herein may result from close contact with dogs and cats observed in all indigenous communities sampled.

45: MICRORNAS AS POTENTIAL BIOMARKERS FOR ARRHYTHMOGENIC RIGHT VENTRICULAR CARDIOMYOPATHY IN BOXER DOGS

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Background: Arrhythmogenic right ventricular cardiomyopathy (ARVC) is a hereditary myocardial disorder characterized by the pathological replacement of myocardial tissue with adipose and fibrous tissue. While prevalent in Boxer dogs, ARVC also affects humans, cats, and other dog breeds, resulting in severe clinical consequences such as malignant ventricular arrhythmias, cardiac dilation, syncope, and sudden death. Diagnosing ARVC often requires invasive histopathological examinations, primarily done postmortem. Given the limitations of current diagnostic criteria, particularly in Boxer dogs, identifying non-invasive biomarkers for early diagnosis and prognosis is imperative. MicroRNAs, small non-coding RNAs that regulate gene expression, are appealing biomarkers due to their stability in tissues and bodily fluids.

Objective: This study aimed to discover microRNA-based biomarkers for the early detection and prognosis of ARVC in Boxer dogs.

Material and Methods: Total RNA was extracted from formalin-fixed, paraffin-embedded cardiac tissue of Boxer dogs affected by ARVC, including those with sudden cardiac death (SCD group; n=4), non-cardiac related deaths (NCD group; n=4), and healthy mixed-breed controls that were euthanized as part of a prior study (CTRL group; n=4). These samples underwent small RNA sequencing (sRNA-Seq).

Results: Small RNA-Seq identified 57 differently expressed microRNAs (fold change ≥ 1.5 ; FDR <0.01) between the SCD and CTRL groups, with only miR-208a downregulated in SCD versus NCRD.

Conclusion: These unique microRNA expression patterns are promising as diagnostic and prognostic markers for ARVC in Boxer dogs. Validation and testing in larger cohorts and in liquid biopsy samples will help further elucidate the potential of microRNAs as diagnostic and prognostic tools.

