Joint Pathology Center
Veterinary Pathology Services

WEDNESDAY SLIDE CONFERENCE 2017-2018

Conference 1

23 August 2017

CASE I: 11-1259-7 (JPC 4017218).

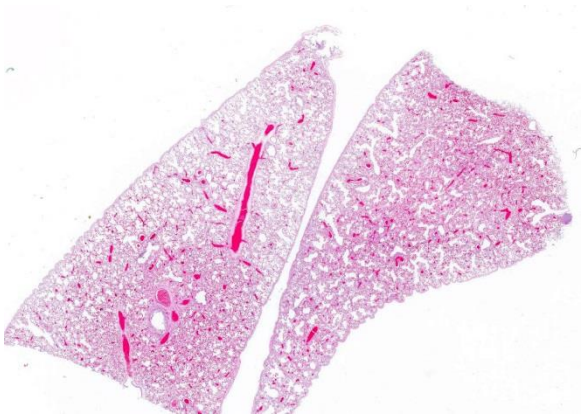
Signalment: Ten-year-old, castrated male, mixed breed canine (*Canis lupis familiaris*).

History: A reportedly 10 year old male castrated mixed breed dog presented for complete necropsy following euthanasia. The dog had a three week history of lethargy, weakness, dyspnea, tachypnea and in-appetence. There was a history of a left total hip replacement and bilateral hip dysplasia. The dog had been on Deramaxx since 2003 and Tramadol since 2009. On presentation the dog was in respiratory distress with a heart rate of 180 bpm. There was pitting edema in the right front forelimb.

Gross Pathology: The dog had a body condition score of 5/5 and was in good post mortem condition. A firm, red, 1cm diameter mass was present on the dorsal antibrachium, and a soft, 7cm x 4cm white subcutaneous mass was present in the right axillary region (lipoma). The pleural cavity contained approximately 150ml of thin red fluid. Within the cranial mediastinum there was a 30 cm x 26 cm by 15 cm lobulated, poorly demarcated, white to yellow firm mass. On cut surface >75% of the mass was soft and yellow to light green (necrosis). The mass extended into the pericardium and similar masses were present within the right and left atria and auricles, and right ventricular free wall (ranging from 3-6cm in diameter). A single 2-3 cm diameter mass was present within the right cranial lung lobe, and another single 2-3 cm mass was present within the wall of the esophagus. There were multiple masses (1-4cm in diameter) within the wall of the stomach.

Gross Morphologic Diagnosis: None provided.

Laboratory results: None provided.

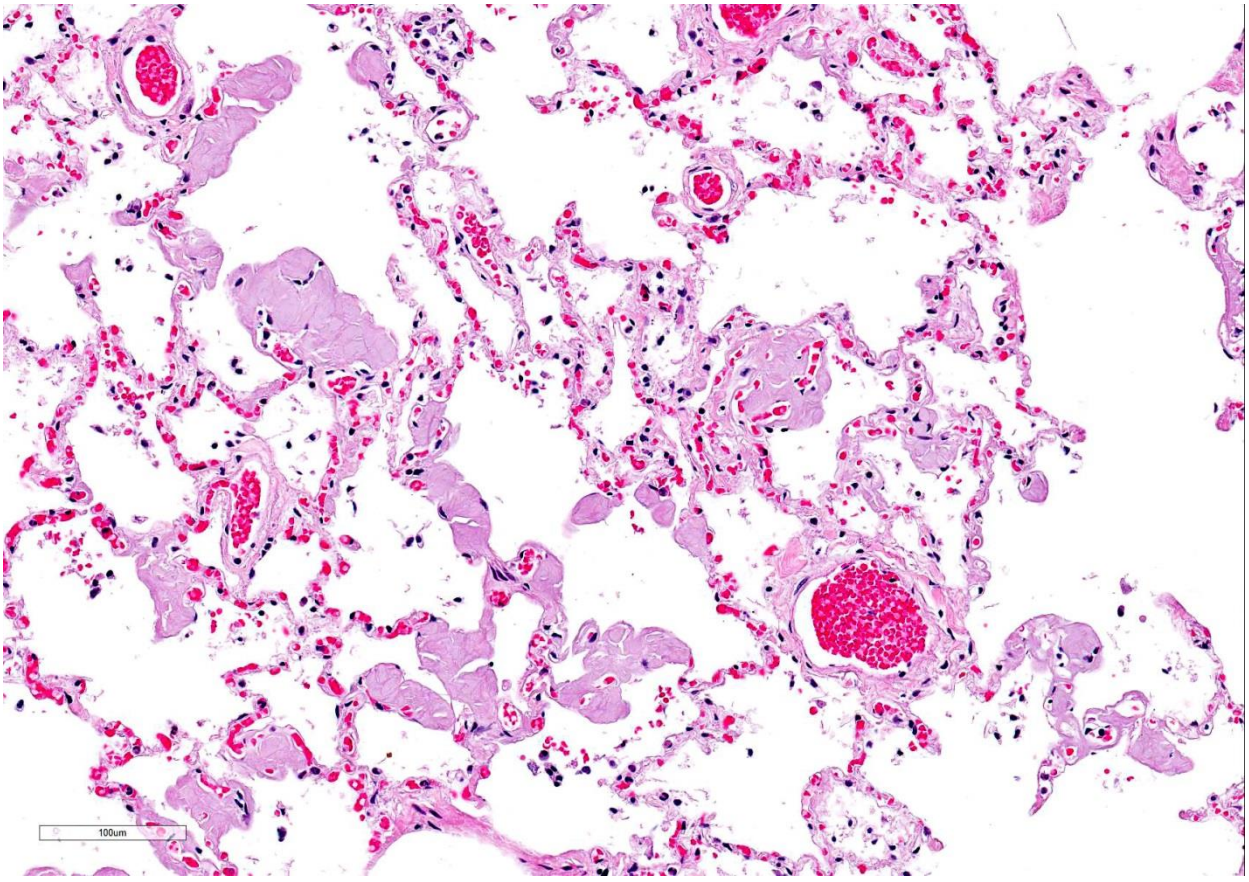


Lung, dog. Two sections are presented for evaluation.

Microscopic Description: Lung: The alveolar interstitium is randomly effaced and expanded by variably sized aggregates of amorphous homogenous eosinophilic material. In addition, similar aggregates are observed replacing the tunica media of pulmonary arteries and arterioles and occasionally of veins. The material can be also seen rarely ruptured into the alveolar air space. Occasionally, organized accumulations of fibrin, neutrophils and erythrocytes are present in arteries and arterioles which are adhered to the endothelial surface of the vessel (thrombosis). Some thrombi contain a flattened cell population adhered to the surface (endothelialization). A moderate multifocal alveolar histiocytosis is present characterized by intra-alveolar accumulations of macrophages with abundant eosinophilic vacuolated cytoplasm. Extramedullary hematopoiesis is present.

Contributor's Morphologic Diagnoses: Marked multifocal pulmonary amyloidosis characterized by deposition in arterioles and interstitium with fibrin thrombi, and mild to moderate pulmonary edema.

Contributor's Comment: Amyloid is composed insoluble protein arranged in β-pleated sheets of protein. The conformation of the protein is the primary characteristic responsible for the binding and staining of the Congo red dye. Amyloid is most commonly classified as secondary or AA

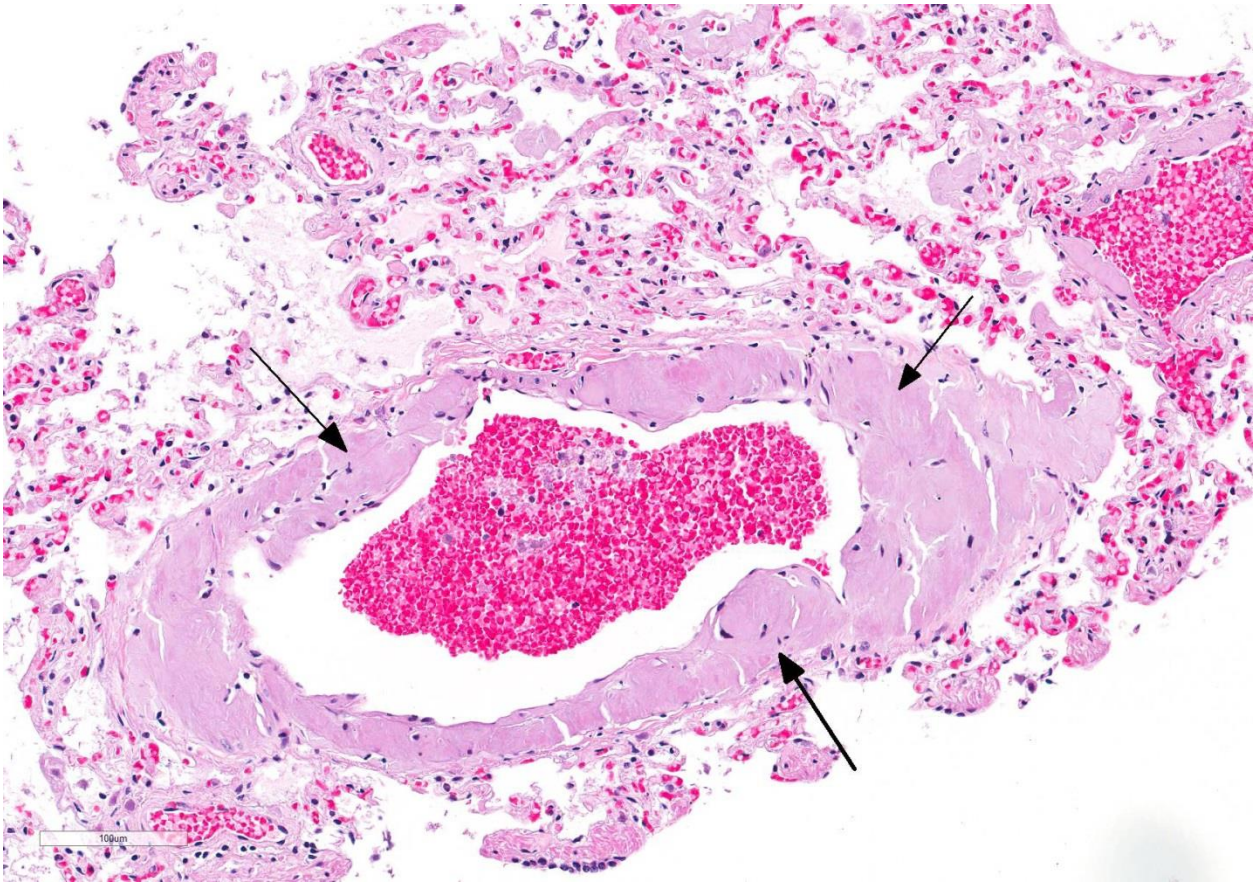


Lung, dog. Two sections are presented for evaluation. Multifocally, nodular aggregates of amyloid expand alveolar septa. (HE, 288X)

and primary or AL in veterinary medicine. AA amyloid is associated with chronic inflammation and is composed of fragments derived from serum amyloid A, a serum apolipoprotein and acute phase protein produced by hepatocytes. Familial amyloidosis in the Shar-Pei dog breed and cat breeds such as the Abyssinian and Siamese is also AA amyloid. AL amyloid is formed from immunoglobulin light chains, predominantly λ light chain fragments. This type of amyloid is commonly associated with plasma cell dyscrasias. Aβ (β-amyloid) is angiocentric cerebral deposits of amyloid recognized in the human and canine. Amyloidosis is also classified as localized or systemic with systemic involvement representing more than 60% of cases (1).

Amyloidosis is reported in several organs in species of veterinary importance but most commonly deposits in the glomerular tuft and peritubular interstitium of the kidney, the periarteriolar lymphoid sheaths in the spleen and the space of Disse in the liver.

Pulmonary amyloidosis is reported in canines and humans. Radiographically, human parenchymal amyloidosis is divided into a nodular form and a diffuse septal form (1). In canines, amyloid deposition in the tunica intima and media of large pulmonary arteries is described. This particular deposition of amyloid is derived from apolipoprotein AI and may be a age related change (2,3). The type of amyloid present in this case was not determined.



Lung, dog. Walls of pulmonary arterioles of all sizes are thickened by amyloid deposits (arrows)(HE, 288X) .

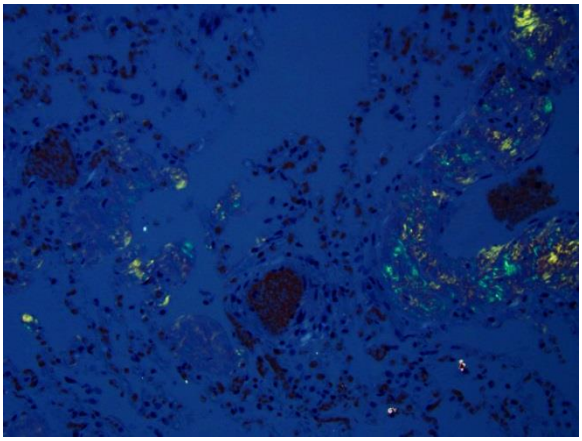
JPC Diagnosis: Lung: Amyloidosis, arteriolar, arterial, and interstitial, multifocal, marked with alveolar edema, mixed breed, *canine*.

Conference Comment: This case nicely demonstrates the histologic changes associated with pulmonary amyloidosis. A Congo red was run to highlight the apple green birefringence of the deposited amyloid and participants discussed the use of Thioflavine T viewed under a ultraviolet light as an additional amyloid marker.[3] Although the microscopic appearance is characteristic, a differential diagnosis of pulmonary arteriolar/arterial and interstitial hyalinosis was discussed. Pulmonary hyalinosis has been reported as a finding in the pulmonary artery of older dogs.[10] Hyalinosis can be differentiated from amyloid using periodic-acid shift (PAS) stain which highlights the glassy eosinophilic material.[2] The following chart was used to review the classification of different types of amyloidosis:

Clinicopathologic Category	Associated Diseases	Major Fibril Protein	Precursor Protein
Systemic (Generalized Amyloidosis)			
Primary Amyloidosis (Immune dyscrasias)	Monoclonal plasma cell proliferations	AL (Light chain amyloid)	Ig light chains (mainly λ type, but also κ type)
Secondary Amyloidosis (Reactive systemic amyloidosis)	Chronic inflammatory conditions	AA (Amyloid associated)	SAA (Serum associated amyloid)
Hereditary Amyloidosis			
Familial amyloidosis	Renal impairment	AA	SAA
Localized Amyloidosis			
Islet amyloid	Type 2 diabetes mellitus	AIAPP	Islet amyloid polypeptide
Cerebral (senile) amyloid	Cognitive disorder	Aβ (Beta-amyloid protein)	APP (amyloid precursor protein)
Pulmonary vessel amyloid	?	Apolipoprotein A-I (apoA-I)	?

Chart adapted from table 6-17 in *Robbins and Cotran Pathologic Basis of Disease*, page 258

While reviewing the chart above, participants were reminded that familial amyloidosis occurs in Shar-Pei dogs and Abyssinian cats with AA amyloid deposition in the renal interstitium as opposed to the glomeruli. In these cases, renal impairment is generally mild and amyloid is usually



Lung, dog. Amyloid deposits within septa and arteriole walls exhibits congophilia and apple-green birefringence on a 5um section . (Congo Red, 400X)

diagnosed incidentally during necropsy.[7]

In addition, conference participants discussed the gross appearance of organs containing amyloid deposits as yellow, waxy, coalescing, nodular amorphous deposits that turns brown when stained with Lugol's iodine and deep purple with acetic acid.[7]

Finally, the conference moderator emphasized (as did the contributor's in the above comment) that in most domestic species AA amyloid deposition in the liver begins in the space of Disse with the exception of the mouse where deposition occurs in the periportal regions of the liver.[1]

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http://vet.osu.edu/biosciences

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CASE II: MU1165314 (JPC 4065317).

Signalment: Two and a half-year-old, female, Charolais bovine, (*Bos taurus*).

History: This cow had a 1 month history of waxing and waning fever, malaise and nasal discharge, with epiphora and bilateral corneal opacity. Clinical signs regressed with dexamethasone treatment. She progressed to sloughing of the skin on the nose, teats, anus, vulva and coronary bands. She was euthanized due to quality of life issues. There was no history of contact with sheep.

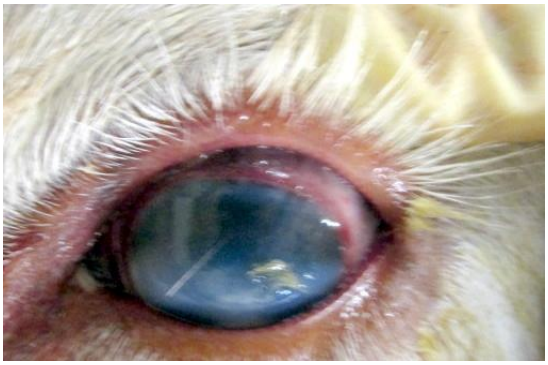
Gross Pathology: This animal was an adequately fleshed, minimally autolyzed white adult female bovine of 500 Kg body weight. The nasal planum was crusted and ulcerated, with red underlying tissue. There was separation of the coronary bands that affected all coronary bands. All teats are covered by crusts, revealing red tissue beneath. Externally both corneas are cloudy and mottled, with reddening of the conjunctiva and milky fluid in the anterior chambers. The corneas became cloudy after fixation and sections of the eye revealed severely increased corneal thickening and exudate in the anterior chamber and behind the lens. The vitreous was cloudy as well.

Lymph nodes associated with the mammary gland, head, neck, and thorax were enlarged to 3-5 times expected volume. There were oral ulcers, particularly on the sides of the thickest part of the tongue and little mucosa remains on the dental pad. The anterior third of the esophagus was uniformly dark red and the wall approached 1 cm in thickness. The abomasal mucosa in 1-1.5 cm thick and the abomasal mucosal folds are thereby accentuated. Punctate ulcers were evident in the mucosa.

Incision of the fixed globes revealed a thickened cornea, with rust red areas of vascularization, and coagulation of exudates in the anterior chamber and vitreous, causing their partial to complete opacity.

Gross Morphologic Diagnosis: None provided.

Laboratory results: Multiple tissues and swabs were positive for herpesviral sequences that were identified as sheep-associated malignant catarrhal fever virus by sequencing. The same samples were negative for sequences of infectious bovine rhinotracheitis virus, bluetongue, BVD and epizootic hemorrhagic disease virus. NVSL

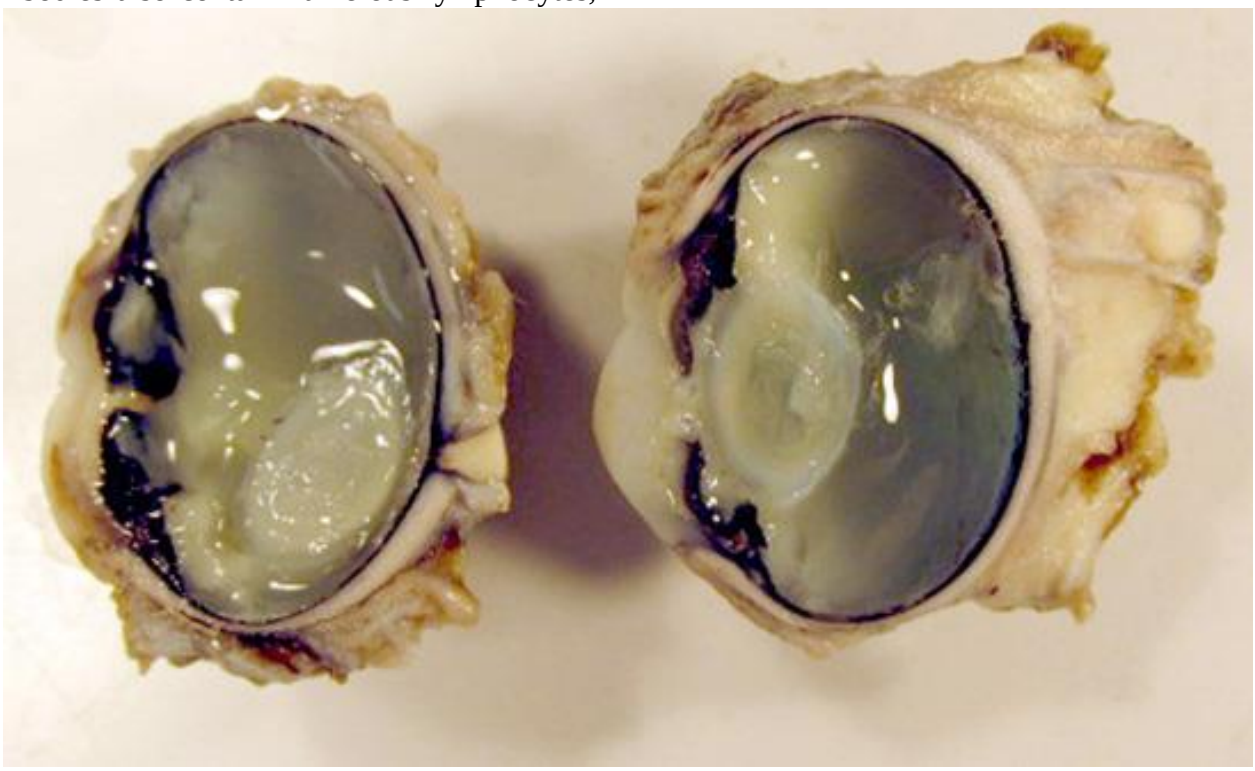


Eye, ox. The cornea is cloudy and edematous, with reddening of the conjunctivae and milky fluid in the anterior chamber. (Photo courtesy of: Veterinary Medical Diagnostic Laboratory, University of Missouri; vmdl.missouri.edu)

testing was declared negative for foot and mouth disease virus.

Microscopic Description (limited to the eye): Nearly every segment of the eye is inflamed or secondarily altered in this animal, with variability in the severity of inflammation between sites . Pink fibrillar edema fluid is present in the anterior chamber. There is pronounced edema of the corneal stroma, with attenuation, vacuolation and loss of the keratinocytes. Intense mixed, predominantly lymphocytic infiltration occurs in the limbus and extends into the cornea, as well as the conjunctiva and sclera. Small thin-walled blood vessels occur in the peripheral corneal stroma, and, beyond this, single file leukocytes align along the stromal fibers. Neutrophils contribute substantially to the population in the more central cornea. The iris and ciliary bodies also contain numerous lymphocytes, macrophages and intermixed neutrophils that exfoliate freely into the anterior and are adhered to the endothelial layer at the back of the cornea. The filtration angle is also filled with similar cells. The fibers of the vitreous are separated by fluid and leukocytes and there choroid is similarly affected. Scleral vessels and extraocular muscle and adventitia have less extensive infiltrates. Lymphocytes are visible in the walls of a few muscular vessels at the base of the iris in some sections.

Similar perivascular lesions (not shown) were associated with ulcerations were found in the skin, tongue, abomasum and in the brain, lung, kidney, heart and adrenal. Lymph nodes were enlarged, with hyperplastic cortical tissue and hemorrhages.



Globe, ox. The cornea is thickened and vascular, and there is coagulated exudate in the anterior and posterior changes. (Photo courtesy of: Veterinary Medical Diagnostic Laboratory, University of Missouri; vmdl.missouri.edu)

Contributor's Morphologic Diagnoses: Eye: Severe lymphocytic vasculitis and perivasculitis, uvea and cornea, with corneal edema, erosion and vascularization.

Contributor's Comment: Malignant catarrhal fever is caused by a rhadinovirus that cause polysystemic disease of cattle, bison, various deer and other ruminants (2). Most cases in cattle affect animals in the 8-24 month age range and have a mean duration of 71 days. Cattle surviving acute MCF have chronic lesions in medium caliber vessel and cornea, comprised of arteriopathy with variable recanalization. Microscopic lesions throughout the body are characterized microscopically by vasculitis. Anterior and posterior synechiae, edema and eventual fibrosis of the corneal stroma, and perforating ulcers and staphyloma are other common ocular lesions.

MCF-related rhadinoviruses have now known to be extremely variable in genetic sequences (3). OvHV-2 had some alleles that varied over 60% in genetic composition.

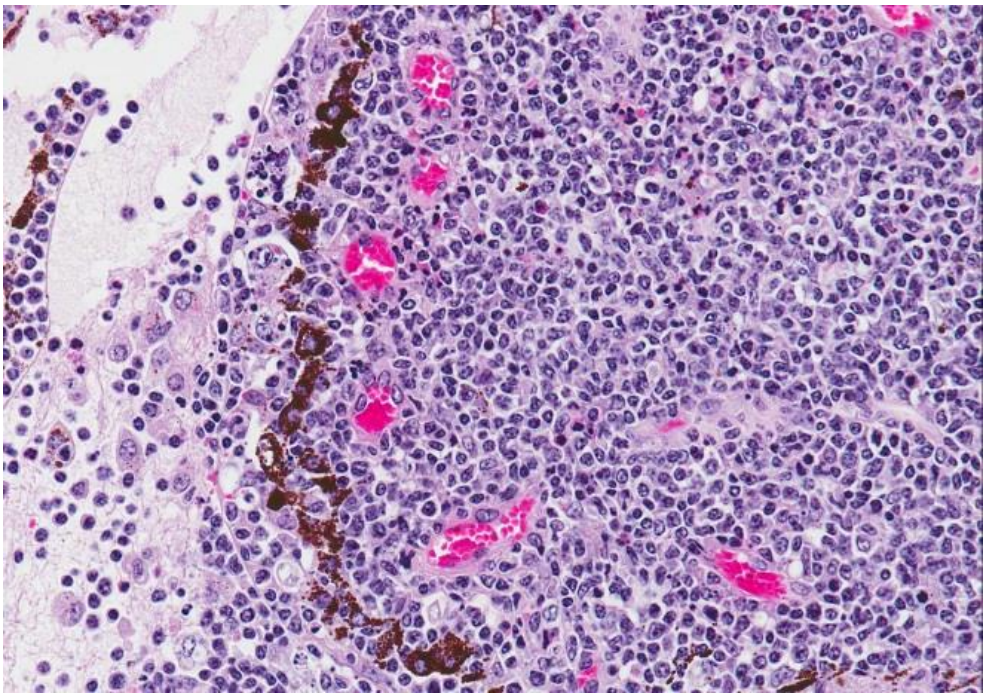
Globe, ox. Subgross examination of a partial section through the affected globe demonstrates an edematous thickened cornea, prominent cellular infiltrate in the uvea, ciliary body and iris leaflets, and proteinaceous exudates within the anterior segment (HE, 50X)

Translation of 9.5 polypeptides revealed only 49% amino acid identity. However, the clinical signs of MCF in cattle from viral isolates originating sheep, bison, reindeer and cattle, as related to viral genotype, did not reveal differences.

Ocular disease is consistently present in the head and eye form but milder lesions can occur in other forms as well (6). In one study, there was no correlation between the degree of corneal edema at first examination and lethal disease outcome. Corneal edema began at the limbus in natural cases, and corneal erosion was common (5). Keratinization of the corneal epithelium, pyknosis and cytoplasmic vacuolation of epithelial cells were observed (4). Corneal perforations occurred and chronic scars common. The corneal edema and uveitis improved in all surviving cattle. Posterior segment disease was frequently present, but difficult to detect to the alterations in the anterior segment (6).

A review of lesions spontaneously occurring MCF-like disease in exotic hooved stock involved cases in 15 moose, 1 roe deer and 1 red deer. Frequent gross findings involved the eye and included conjunctivitis, corneal opacity and fibrin clots in the anterior chamber. Although OvHV-2 caused some cases, CpHV-2 caused others. Most cases occurred in farmed animals and zoos (1). The microscopic appearance of lesions was similar to those in cattle. Additional novel rhadinoviruses have been described in exotic hooved stock and cervids. (1) Pigs also develop ocular lesions resulting from MCF (2).

JPC Diagnosis: Eye: Panuveitis and vasculitis, lymphoblastic and necrotizing, diffuse, severe with ulcerative keratitis and corneal edema, Charolais, bovine.



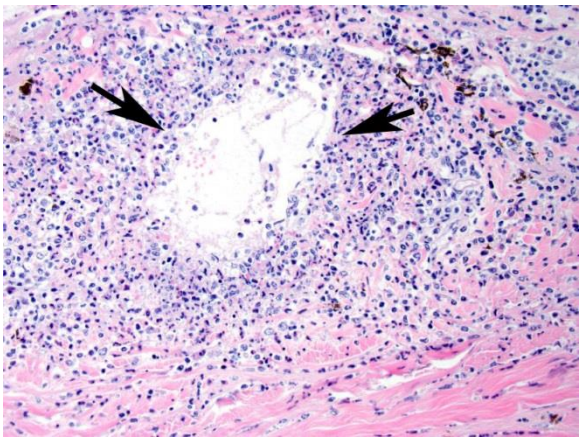
Globe, ox. The ciliary body is markedly expanded by an infiltrate of large numbers of blastic lymphocytes admixed with fewer heterophils and cellular debris. (HE 400X)

Conference Comment: There was significant slide variation in this case. Additional morphologic diagnoses generated by conference participants included conjunctivitis, keratitis, and episcleritis depending on the plane of tissue sectioned.

Malignant catarrhal fever is caused by infections with the MCF virus group of ruminant *Gammaherpesviridae* (which are known as Rhadinoviruses in older texts).[3] Of the pathogens in the MCF virus group, 6 are associated with clinical signs: *Alcelaphine herpesvirus* 1 (carried by wildebeest) and 2 (carried by hartebeest), *Ovine herpesvirus* 2 which is endemic in domestic sheep, *Caprine herpesviruses* 2 (endemic in domestic goats) and 3 (affects white tailed deer and red brocket deer), and *Ibex MCF virus* which is carried by Nubian ibex and produces disease in bongo and anoa. [5] However, most natural outbreaks are due to *Ovine herpesvirus* 2 in sheep or *Alcelaphine herpesvirus* 1 in African wildebeest.

MCF is characterized by marked T-lymphocyte hyperplasia which was prominent in conference discussion. [5] In the slides examined numerous lymphoblastic cells were present in various portions of the eye with prominent mitotic figures. The pathogenesis of MCF is presumed to start with infection of large granular lymphocytes that are subsequently transformed by the gammaherpesvirus. In fact, the OHV-2 genome has been detected in CD8+ T cells which are the predominant cell present in the perivascular inflammation. The pathogenesis of this disease is unclear, and although the invasive T cells are most likely cytotoxic T lymphocytes or T-suppressor cells the mechanism they use to cause such marked vasculitis has not yet been identified.[5]

Conference participants briefly reviewed various terms used to classify ocular inflammation such as: endophthalmitis (inflammation of the uvea, retina, and ocular cavities), panophthalmitis (inflammation of all of the ocular structures, including the sclera), anterior uveitis (inflammation of the ciliary body and iris), posterior uveitis (inflammation of the ciliary body and



Globe, ox. The wall of a scleral venule is expanded and effaced by infiltration of blastic lymphocytes and heterophils which are admixed with cellular debris (vasculitis). (HE 400X)

choroid), panuveitis (inflammation of the iris, ciliary body, and choroid), and chorioretinitis (inflammation of the choroid and the retina).[7]

In addition, these slides contained nice examples of the tapetum lucidum which is not commonly seen in microscopic sections. The conference moderator noted that in cats and dogs the tapetum is cellular and has a "brick-like" appearance; whereas in ruminants and horses, it contains more fibrous connective tissue with fibroblasts arranged linearly. Pigs were specifically mentioned because they are lacking a tapetum lucidum.[1]

Acute severe bovine viral diarrhea (BVD) and mucosal disease was mentioned as a differential. However, MCF usually affects multiple organs that are not involved in mucosal disease like liver, kidney, bladder, eye, and brain. Also, MCF produces lymphoid hyperplasia whereas lymphoid tissue in BVDV infections is atrophic.[5]

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CASE III: RP22064 (JPC 4066355).

Signalment: Adult, female, brush rabbit (*Sylvilagus bachmani*).

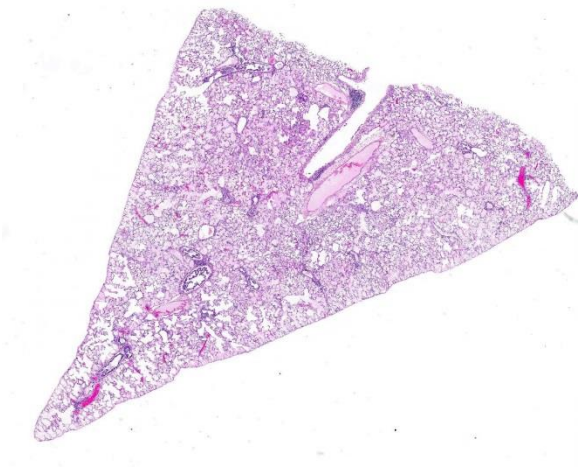
History: This animal was 1 of 2 rabbits found dead in a lion exhibit in the same week.

Gross Pathology: The rabbit had small adipose stores. There were fleas in the haircoat. The lungs were red but floated in formalin.

Gross Morphologic Diagnosis: None provided.

Laboratory results: *Toxoplasma gondii* was confirmed by PCR and sequencing performed on tissues from the second brush rabbit found in the same enclosure.

Microscopic Description: Examined is a section of lung in which alveolar lumina are diffusely flooded with proteinaceous fluid (edema) and beaded eosinophilic fibrillar material (fibrin) admixed with plump and foamy alveolar macrophages, fewer lymphocytes, nondegenerate heterophils and plasma cells and scant hemorrhage. Small amounts of fibrin and few lymphocytes, plasma cells, heterophils, and macrophages thicken alveolar septa (up to 3 times normal thickness) and surround larger pulmonary blood vessels. Within alveolar septa and lumina, there are moderate numbers of 1 - 2 µm, oval to fusiform basophilic organisms (tachyzoites) forming 15 - 30 µm in diameter clusters (presumed intrahistiocytic) or, less often, arranged individually (extracellular). Multifocally, there is scattered lytic necrosis of alveolar septa characterized by disruption and replacement of septa by small amounts of necrotic cellular and karyorrhectic debris, fibrin and the aforementioned inflammatory cells. Bronchiole lumina contain refluxed edema fluid and inflammatory cells and peribronchiolar connective tissue is edematous. Alveolar septa are rarely lined by a thin layer of brightly eosinophilic fibrin (hyaline membranes).



Lung, rabbit. At subgross magnification, there are multiple randomly scattered foci of hypercellularity and multifocal to coalescing areas of edema-filled alveoli. (HE, 5X)

Contributor's Morphologic Diagnoses: Lung: Moderate diffuse acute interstitial pneumonia with edema, necrosis, and intralesional protozoa (etiology: *Toxoplasma gondii*).

Contributor's Comment: Histologic findings are consistent with *Toxoplasma gondii*.[2,3] *T. gondii* are apicomplexan protozoa that affect a wide range of intermediate hosts and are closely related to other coccidia including *Neospora* spp. and *Sarcocystis* spp.[2]

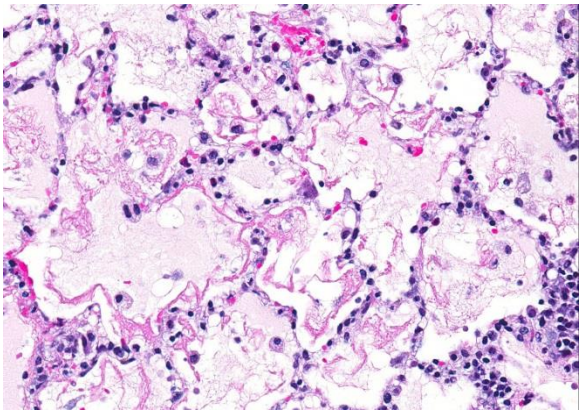
The life cycle of *T. gondii* includes both an intermediate and definitive host. All warm-blooded mammals, including cats and humans, can act as intermediate hosts to complete the asexual stages of the life cycle.

The intermediate host is infected by ingesting the sporulated oocysts in food, water, or soil contaminated with cat feces, or less commonly, by ingesting tissue cysts in uncooked meat.[1,3] Sporozoites leave the oocyst and develop into tachyzoites, which invade the lamina propria and multiply in the intestines. Tachyzoites continue to multiply first in the mesenteric lymph nodes, and then reach the rest of the body through the circulation either free or within lymphocytes, macrophages, and granulocytes. Lesions associated with acute toxoplasmosis are more commonly observed during this asexual stage with rapid multiplication of tachyzoites. The most commonly associated lesions include necrosis, edema, and inflammation (Figure 1).[1-3] This case shows the lesions of an acute infection with *T. gondii*. In this rabbit, lesions were observed in the lung, heart, liver, spleen, kidney, adrenal gland, thyroid gland, stomach, small intestine, brain, skeletal muscle, mesenteric lymph node, and nasal turbinates. Tachyzoites can spread anywhere in the body, as evidenced in this case. Tachyzoites eventually encyst in a wide range of tissues, including brain, liver, lung, muscles, and retina. Tissue cysts may contain anywhere from two to hundreds of bradyzoites. Animals that survive the acute phase of the infection acquire immunity to *T. gondii*.[2]

Domestic and wild felids are the definitive hosts for *T. gondii*. Cats most commonly become infected by eating muscle containing tissue cysts. Bradyzoites are released in the gastrointestinal tract and enter the epithelial cells of the small intestines to undergo several stages of asexual and sexual multiplication. Gamonts are formed and result in oocyst formation. The prepatent period is variable and ranges from 3 – 18 days or more depending on the stage of the organism at the time of ingestion. Unsporulated oocysts are released into the feces where they sporulate within 24 hours in order to infect an intermediate host. Oocysts can remain viable in the environment for long periods of time.[1,2]

This case shows the lesions of disseminated toxoplasmosis with a rabbit as the intermediate host; the definitive host in this case is unknown. The lions may have functioned as the definitive host, or the rabbits could have acquired the infection outside of the lion enclosure. Biosecurity measures are in place to exclude feral cats and other felids from the entire facility. In general, biosecurity measures that could decrease the impact of *T. gondii* in zoos and other facilities include: not housing highly susceptible species (e.g. marsupials and primates) near felids, freezing meat that cannot be cooked prior to feeding animals, designing enclosures to exclude domestic cats and other vectors, and daily cleaning to prevent the sporulation of oocysts in the environment.[2]

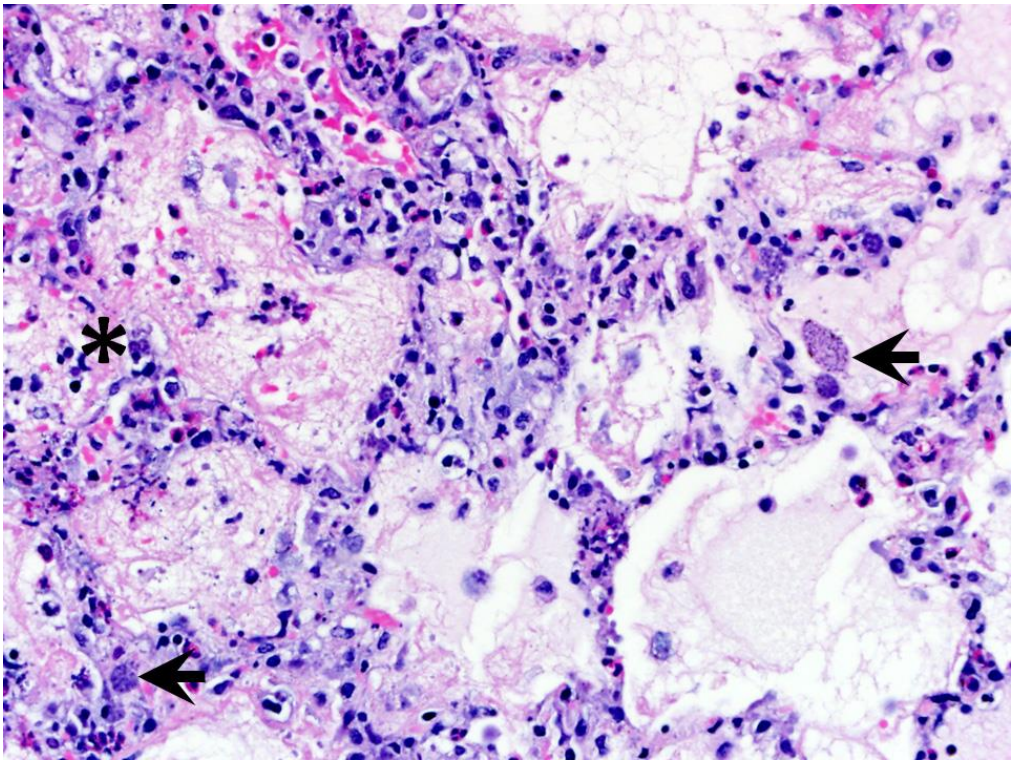
JPC Diagnosis: Lung: Interstitial pneumonia, necrotizing, diffuse, moderate, with fibrin, edema, and protozoal tachyzoites (etiology consistent with



Lung, rabbit. Alveoli are flooded with edema fluid and polymerized fibrin, which often forms hyaline membranes long hypercellular, and often necrotic alveolar walls. (HE, 320X)

Toxoplasma gondii), brush rabbit, *Sylvilagus bachmani*.

Conference Comment: This case nicely demonstrates the characteristic histologic lesions associated with the interstitial pneumonia associated with systemic toxoplasmosis. Conference participants described the random areas of necrosis and identified intracellular and extracellular protozoal tachyzoites within the interstitium and epithelial cells. Participants discussed the different patterns of pneumonia, interstitial, bronchopneumonia, embolic pneumonia, bronchointerstitial pneumonia, granulomatous, and uncategorizable pneumonias.



Lung, rabbit. Several intracellular and extracellular clusters of tachyzoites (arrows) are present within alveolar macrophages and alveolar septa. Tachyzoites are 1 - 2 microns in diameter with a basophilic nucleus. There is necrosis of alveolar septa with associated lymphohistiocytic and heterophilic infiltrates, fibrin, and karyorrhectic debris (asterisk). (HE, 400X) (Photo courtesy of: Wildlife Disease Laboratories, Institute for Conservation Research, San Diego Zoo Global, http://www.sandiegozooglobal.org)

The conference moderator discussed traditional bronchopneumonia, which involves an exudate originating at the bronchiolar-alveolar junction and fills bronchioles and alveoli. The characteristic distribution is cranioventral (due to gravity dispersal of inhaled pathgens) which is most commonly caused by opportunists. In contrast, bronchointerstitial pneumonia may be defined as either (1) bronchiolar necrosis and diffuse alveolar damage with destruction of both bronchiolar and alveolar epithelium, or (2) mononuclear cellular inflammation which surround airways and infiltrate alveolar septa. *Mycoplasma hyopneumonia* with its characteristic peribronchiolar and peribronchar lymphoproliferative nature was discussed as one example.[4]

Conference participants considered *Encephalitozoon cuniculi* as a potential differential for this presentation in a rabbit. *Encephalitozoon cuniculi* is a gram-positive, acid fast, obligate intracellular microsporidian that infects a variety of mammalian hosts, the domestic rabbit, being one of the most common. Historically, there has been disagreement among taxonomists about the

classification of this organism, but genomic sequencing has confirmed it as a eukaryotic fungus within the phylum Microsporidia. Transmission occurs through ingestion or inhalation of infected urine or transplacentally. Infective spores then enter circulation via infected mononuclear cells and hit initial target organs such as lung, liver, or kidney. At approximately 3 months post infection, organisms can be found within the central nervous system and produce characteristic clinical signs (head tilt, ataxia, and vestibular signs). In dwarf rabbits, phaecoclastic uveitis and cataract formation are quite common after transplacental transmission. In the lung, lesions may appear as a focal to diffuse interstitial pneumonia with mononuclear cellular infiltration. Microscopically, *E. cuniculi* and *T. gondii* can look quite similar; staining characteristics can help differentiate the two. Toxoplasma organisms are Gram-negative and do not stain with carbol fuschin stains (a type of acid fast stain, a Ziehl-Neelsen subcomponent stain).[2] Although, the contributor prudently identified toxoplasmosis using PCR, Gram stains and several acid fast stains (Ziehl-Neelsen and Fite-Faraco; we do not have access to Carbol fuschin) were performed to further rule out encephalitozoonosis. Organisms within submitted sections were strongly Gram negative, and were not highlighted with acid fast stains which confirm the contributor's diagnosis of *Toxoplasma gondii*.

Finally, conference participants discussed that Toxoplasmosis has a military and veterinary public health relevance due to its recent identification in the central nervous system of individuals suffering from various mental health disorders (bipolar disorder, post-traumatic stress disorder, schizophrenia) and related suicides.[1]

Toxoplasma	***Encephalitozoon***
Small cyst 60	Large pseudocyst up to 120
Spores not acid fast	Spores are **acid fast**
Gram negative	Gram **positive**
Do not stain with carbol fuchsin	Stain with **carbol fuchsin** (purple)
Giemsa: granulated cytoplasm	**Giemsa**: light blue cytoplasm
Stains well with **H&E**	Stains poorly with H&E
Larger organism 2-6 um	Smaller organism 1.5 x 2.5 um
Tend to invoke **necrosis**	Necrosis is not a feature

Contributing Institution:
Wildlife Disease Laboratories
Institute for Conservation Research

San Diego Zoo Global
http://www.sandiegozooglobal.org

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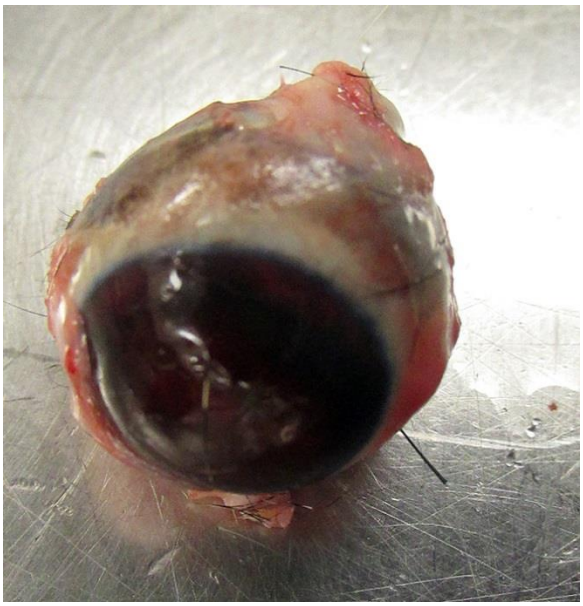
CASE IV: UMC171 (JPC 4099789).

Signalment: Sixteen-year-old, neutered male, Domestic shorthair cat (*Felis catus*).

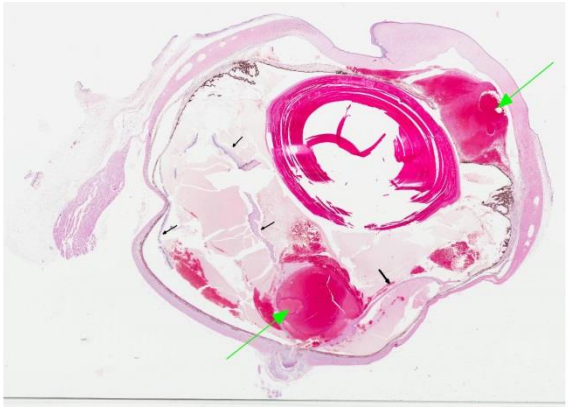
History: The cat had an enlarged thyroid discovered during dental prophylaxis 1.5 years before death. At this time there was a grade I murmur auscultated over the left heart. Anti-thyroid medication was started but was discontinued due to miscommunication with the owner about the need for life-long therapy. Four months before euthanasia the cat was noted to be losing weight and was presented again. It was treated for hyperthyroidism and hypertension. Three weeks before euthanasia, the cat became depressed and painful over several days and was brought to an emergency service. He was found to have hyphema in both eyes, with elevated intraocular pressure in the left eye. BUN was also increased. After several days of trying unsuccessful medical therapy and pain relief, the cat was euthanized. The cardiac murmur had become grade IV of VI at three days before euthanasia.



Globe, cat: Gross view of the left eye. The anterior chamber is filled with blood and an ulcer is present near the lateral cornea. (Photo courtesy of: Veterinary Medical Diagnostic Laboratory, University of Missouri)

Gross Pathology: An aged, neutered male, silver tabby and white, short-haired feline is necropsied. The animal has body weight 4.6 kg, with adequate fat stores and minimal

autolysis. The left eye is filled with blood, and the cornea bulges forward. An ulcer covers the central cornea. Blood leaks into the fixative when the eye is immersed in formalin after removal. The left thyroid is dark brown in color and enlarged, with a length of 1.5 cm. The right thyroid is reduced in size, nodular, and atrophic. The parathyroid glands are prominent. The kidneys are pale tan in color with a slight indentation of the anterior pole of the left kidney. They are firm in texture and the cortex has a somewhat granular character, with reduced cortical width. The combined kidneys weigh 33.5 grams, 0.72% body weight (normal 1.1%).The heart, especially the left ventricle, is severely enlarged. The total heart weight is 25.2 grams, 0.54% body weight. The right ventricular free wall weighs 3.0 grams (11.0% total heart weight) and the left ventricle weight 16.0 grams (63% heart weight, .35% body weight). The left to right ventricular weight ratio is 5.33, increased). The right ventricular wall measures 2 mm in width and the left 11 mm (ratio 5.5, increased).

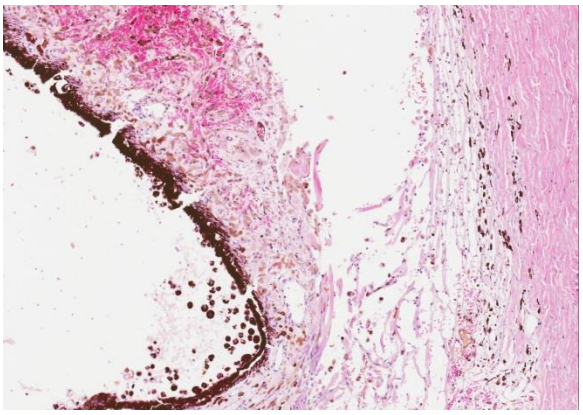


Globe, cat: There are large clots in the anterior and posterior segments (green arrows), and the retina is diffusely detached. (black arrows). (HE, 60X)

Gross Morphologic Diagnosis: None provided.

Microscopic Description: The anterior portion of the eye is filled with hemorrhage, including the angle, with extensive hemosiderosis at the root of the iris and in the meshwork. Additional hemorrhage mixes with the vitreous in parts of the posterior chamber and lies on both sides of a detached retina. The RPE is universally hypertrophic. The detached retina is severely atrophic, with reduction and mixing of the granular layers and with loss of the ganglion cells, particularly at the periphery. Scattered hemosiderophages are also present in the retina and small arterioles are thickened. Adjacent optic nerve contains few axons (not present in all slides). Thick, hyalinized arterioles occur in the choroid and retina. PAS staining highlights increased eosinophilic material in the media of small muscular vessels. A segment of corneal erosion is attended by stromal disarray, melanosis and vascularization, with mild superficial inflammation. A narrow fibrovascular membrane extends along the anterior face of the iris.

Contributor's Morphologic Diagnoses:
Hyphema with glaucoma, retinal detachment, retinal atrophy and corneal erosion with keratitis
Arteriolar degeneration (arteriolosclerosis), eye
Other pertinent final diagnoses: Thyroid adenomas, atrophy of normal thyroid, left ventricular hypertrophy, glomerulosclerosis with similar vascular lesions (not included).



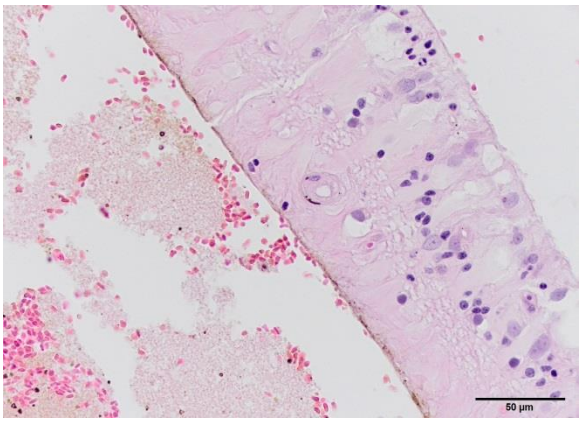
Globe cat. A hemorrhagic pre-iridal fibrovascular membrane covers the iridal root; however, the drainage angle remains open. (HE, 168X)

Contributor's Comment: This particular patient had multiple risk factors for developing hypertensive retinopathy and hyphema. A combination of thyroid-induced ventricular enlargement and renal failure, along with irregular treatment of the condition, produced a cycle of worsening disease. Histologic lesions primarily involve retinal and choroidal vessels, as in this case, with lesions ranging from fibrinoid necrosis to multi-layered onion-skin layering of medial hypertrophy and adventitial fibrosis. The animal has severe separation of the retina from the choroid with choroidal hypertrophy and hemosiderosis. Given the severity of retinal atrophy, it is likely that the cat had been blind for several weeks before hyphema was noticed clinically.

Intraocular hemorrhage is a frequent sequelum of high blood pressure in old cats.[2,5] Renal disease, hyperthyroidism and cardiac disease are commonly contributory (this cat had a trifecta). Disease is usually symmetrical but with qualitative differences between eyes. This cat also developed hyphema in the right eye, but intraocular pressure did not become elevated and no corneal ulcer was found. Hypertensive lesions can be found in the retina, choroid and rarely iris. Vascular lesions result in retinal and preretinal hemorrhage and edema; with retinal detachment because of effusion from leaky choroidal vessels. Exudative retinal separation and retinal necrosis produce atrophy of the photoreceptive and characteristic "tombstoning" of the retinal pigment epithelium. In this eye intraocular pressure was increased, resulting in secondary open angle glaucoma.

The earliest retinal changes in hypertension are arteriolar narrowing secondary to vasospasm, followed by diffuse or focal narrowing of arteriolar walls, changes that can be seen on ocular exam. In progressive disease, the blood-retinal barrier breaks down, leading to fluid leakage, bleeding and ischemia of the nerve fiber layer. Severe disease in people portends an increased risk of cardiovascular mortality.[1]

Hyphema or hemorrhage into the anterior chamber results from disruption of the blood ocular barrier, and has a number of causes, including trauma, vessel-rich neoplasms and coagulopathy. In some of these conditions, only one eye is affected. Common sequelae include cataracts, glaucoma, synechiae, corneal staining by hemoglobin, and eventual phthisis. Other organs, including brain, heart and kidneys are common targets



Globe cat. The detached retina is atrophic, with a few granular neurons remaining. A hyalinized arteriole is visible in the center of the retinal segment. (HE, 400X) (Photo courtesy of: Veterinary Medical Diagnostic Laboratory, University of Missouri)

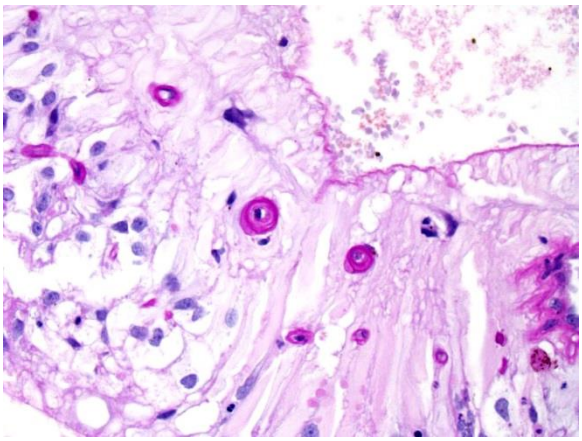
of hypertension. Bleeds due to hypertension can produce clotted or unclotted blood. Use of ultrasound can be useful, if blood obscures evaluation of the back of the eye. Persistent slow bleeding and alteration of intraocular structures suggests a poor prognsis.[4]

Decreased blood flow through the vasa vasorum during hypertension results in acute hypoxia and aortic medial necrosis. In turn, decreased flow results in increased vascular tone, neovascularization, leading to reduced vasodilatory capacity, creating a self-perpetuating cycle. Kidney disease, hyperthyroidism, hyperaldosteronism, anemia and diabetes are predisposing systemic diseases. Some studies indicate that arteriosclerosis is uncommon in normotensive cats. Remodeling of small arteries and arterioles results in narrow lumen diameter and hypertrophic remodeling of the media.[3]

JPC Diagnosis: Eye: Arteriolosclerosis, multifocal, moderate with severe intraocular hemorrhage, retinal atrophy and detachment, Domestic shorthair, feline.

Conference Comment: This case provided exceptionally descriptive microscopic lesions. Intraocular hemorrhage was the most prominent microscopic finding among conference participants. There was dialogue about the top four rule outs for hemorrhage in all chambers of the eye:

(1) Trauma (often a diagnosis of exclusion)
(2) Hypertensive retinopathy
(3) Neoplasia which can rupture and hemorrhage into the eye
(4) Inflammation especially due to hematogenous uveal localization of infectious agents
(5) Coagulopathy



Globe cat. A periodic acid-Schiff stain demonstrates the 'onion-skinning" changes associated with the walls of small vessels in the atrophic retina. (PAS, 400X)

It was obvious amongst conference participants that neoplasia and inflammation could be ruled out. However, since participants were not provided the clinical history in this case, a conversation ensued regarding how to separate trauma from hypertension microscopically.

Vascular changes are the best way to differentiate hypertension from trauma. With systemic hypertension, there is fibrinoid necrosis of the tunica media, thickening of arteriolar walls, narrowing of the vascular lumen most likely secondary to vessel damage and leakage of blood proteins into the wall which is best seen in the choroid and retinal vessels.[6] The thickened walls and characteristic "onion-skinned" appearance were highlighted in this case with a periodic acid-Schiff stain. Based on our microscopic findings, we concur with the contributor's diagnosis of hypertensive retinopathy and discussed the clinical findings in this case (described above) to include the associations of renal failure and hypertension/elevated intraocular pressure.

Renal disease is the most common cause of hypertension in dogs and cats and may

either be a cause or an effect of hypertension. In either case, hypertension is self-perpetuating because medial hypertrophy and hyalinization of renal arteries lead to progressive nephrosclerosis, heightened hypertension, and increased pressure-induced damage in affected tissues.[4] In this case, it is not clear which was the initial development: hyperthyroidism, renal disease, or hypertrophic cardiomyopathy. However, it is likely that all three contributed to the clinical and microscopic findings in this case.

Additionally, conference participants viewed several iridal changes, resulting in discussion of the following entities (not all present in this slide): anterior synechia (adherence of the iris to the cornea with compression of the drainage angle), posterior synechia (adherence of the iris to the lens), iris bombé (posterior synechia that involves the entire circumference of the iris and blocks flow of aqueous causing increased pressure in the posterior chamber and causing bowing of the iris forward), ectropion uveae (contraction of a pre-iridal fibrovascular membrane resulting in infolding of the pupillary border to adhere to the anterior iris surface), and entropion uveae (contraction of a pre-iridal fibrovascular membrane resulting in infolding of the pupillary border to adhere to the posterior iris surface).[6]

Contributing Institution:
Veterinary Medical Diagnostic Lab and Department of Veterinary Pathobiology
http://vmdl.missouri.edu/
http://vpbio.missouri.edu/

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