

# **50<sup>th</sup> AAVLD Diagnostic Pathology Slide Session**

**American Association of Veterinary Laboratory Diagnosticians**



**Reno, Nevada  
October 20, 2007**

**50<sup>th</sup> AAVLD Diagnostic Pathology Slide Session**  
**October 20, 2007; 3:30-6:00PM**  
**Reno Nevada**

| <b>Case/Page #</b> | <b>Base Slide Label</b> | <b>Presenter</b>   | <b>Species</b> |
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| 1                  | 07-19605                | Jaime Paulin       | canine         |
| 2                  | AAVLD 2007 Case#2       | José A. Ramos-Vara | guinea pig     |
| 3                  | E06-153-2               | C.G. Lamm          | fish           |
| 4                  | #4 91-118-4             | Laura Coffee       | coati          |
| 5                  | Case 5                  | Dodd Sledge        | feline         |
| 6                  | NF-07-871               | Molly Seavey       | porcine        |
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| 8                  | 07011738                | C.G. Lamm          | canine         |
| 9                  | Case 9 Ohio ADDL        | Jeffrey Hayes      | porcine        |
| 10                 | 50408707 13             | F. Uzal            | equine         |
| 11                 | AAVLD 2007 Case#11      | Joshua Webster     | bovine         |
| 12                 | DCPAH Case 12           | M.C. Boyle         | feline         |
| 13                 | AAVLD 2007 Case 13      | Rob Bildfell       | chameleon      |
| 14                 | 50700565 H1             | S. Diab            | bovine         |
| 15                 | F0700831                | H. Shivaprasad     | parrot         |
| 16                 | F0604897                | H. Shivaprasad     | parrot         |
| 17                 | AAVLD 2007 Case #17     | Gayle Johnson      | bovine         |
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| 19                 | AAVLD 2007 Case #19     | Gayle Johnson      | canine         |

Session Coordinator: Gayle C. Johnson, University of Missouri

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| <b>Case/Page #</b> | <b>Presenter</b>   | <b>Species</b> | <b>Diagnosis</b>                 |
|--------------------|--------------------|----------------|----------------------------------|
| 1                  | Jaime Paulin       | canine         | Ciliary dyskinesia               |
| 2                  | José A. Ramos-Vara | guinea pig     | Endometrial hyperplasia          |
| 3                  | C.G. Lamm          | fish           | Lymphocystis, fibrosarcoma       |
| 4                  | Laura Coffee       | coati          | Ceroid lupofuscinosis            |
| 5                  | Dodd Sledge        | feline         | Hepatic amyloidosis              |
| 6.                 | Molly Seavey       | porcine        | Hepatic ascariasis               |
| 7                  | Andy Allen         | equine         | Nocardioform placentitis         |
| 8                  | C.G. Lamm          | canine         | Hypertrophic osteodystrophy      |
| 9                  | Jeffrey Hayes      | porcine        | Selenium intoxication            |
| 10                 | F. Uzal            | equine         | Brainstem mineralization         |
| 11                 | Joshua Webster     | bovine         | Herpesviral enteritis            |
| 12                 | M.C. Boyle         | feline         | Melamine-associated nephropathy  |
| 13                 | Rob Bildfell       | reptile        | Fungal dermatitis/microfilaremia |
| 14                 | S. Diab            | bovine         | Zygomycotic lymphadenitis        |
| 15                 | H. Shivaprasad     | parrot         | Yersinia hepatitis               |
| 16                 | H. Shivaprasad     | parrot         | Renal atherosclerosis            |
| 17                 | Gayle Johnson      | bovine         | Hydranencephaly                  |
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# Primary Ciliary Dyskinesia in a Dog

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University of Minnesota

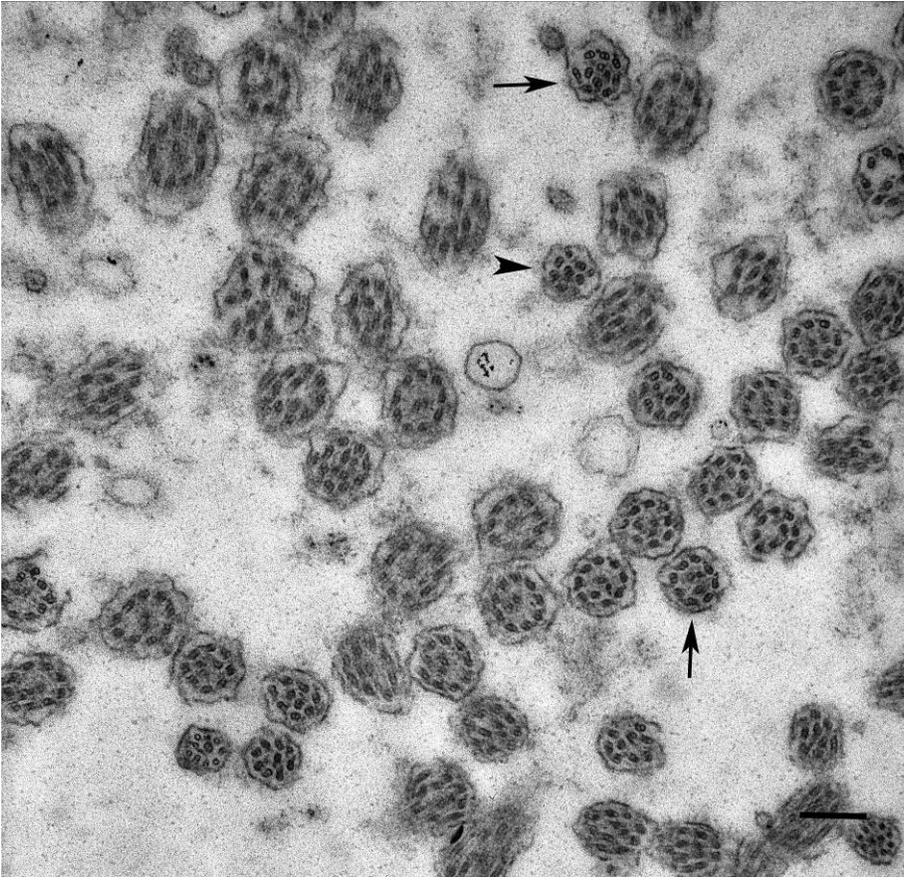
This dog was an 8-month-old, female, mixed breed with a history of chronic respiratory disease characterized by bronchointerstitial pneumonia with secondary bacterial infection. Postmortem gross and histologic findings consisted of diffuse, marked to severe catarrhal-purulent sinusitis and bronchopneumonia with gram negative (predominantly) and gram positive bacteria within the bronchi; multifocal, mild to moderate suppurative tracheitis with hyperplasia of epithelial cells; bilateral, symmetric, moderate internal hydrocephalus; and bilateral, moderate chronic otitis media with abundant ceruminous exudate. Ultrastructurally, 80% of the cilia of the nasal and bronchial epithelium had abnormalities in the microtubule structure (fig.1), including absence of outer and inner dynein arms, disorganization of peripheral doublets, centrally located doublets, and supernumerary singlets. Abnormal microtubule patterns included 6+1 to 6+4, 7+1 to 7+4, 8+1 to 8+3, and 9+0 to 9+2 (doublet+singlet).

## Comments

The structural abnormalities detected by transmission electron microscopy were diagnostic for primary ciliary dyskinesia (PCD). PCD is an autosomal recessive genetic disorder in humans, which occasionally affects animals as well. Generally, PCD patients have persistent and/or recurrent respiratory disease due to impaired mucociliary clearance, and may have neurologic signs secondary to hydrocephalus. Kartagener's syndrome is a subset of PCD which combines bronchiectasis and sinusitis, as well as situs inversus which results from abnormal cilia motility during embryogenesis. Tests used in the diagnosis of PCD include transmission electron microscopy, scintigraphy to evaluate the mucociliary clearance, and ciliogenesis to evaluate ciliary beat synchrony and to rule out secondary ciliary lesions.

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**Figure 1.** Electron micrograph; Cross section of respiratory cilia; Dog, primary ciliary dyskinesia. Multiple microtubules have abnormal doublet+singlet patterns 7+4 and 8+3 (arrows), disorganized peripheral doublets, centrally located doublets (arrowhead) and supernumerary singlets. Bar = 200nm.

## **Cystic adenomatous endometrial hyperplasia and leiomyoma in a guinea pig (*Cavia porcellus*)**

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This 3-year-old guinea pig sow had a 2-year history of an abdominal mass and more recent onset of bilaterally symmetrical alopecia. Exploratory laparotomy revealed a markedly enlarged uterus (body and both horns) and cystic left ovary. Hysterectomy was performed. Grossly, the uterus was diffusely enlarged with average external diameter of 1-2 cm. A 1.3 cm-diameter pedunculated mass protruded from the cranial aspect of the left uterine horn, elevating the serosa. The uterine mucosa was markedly expanded and contained multiple cysts with clear to mucoid fluid. Microscopically, the uterine body and horns had mucosal and glandular epithelial proliferation with cystic dilation of glands. Glands were lined by a variety of epithelial types: simple cuboidal, simple columnar, pseudostratified, keratinizing stratified squamous, and transitional epithelia. In some glands, two different epithelia coexisted, with abrupt transition. The most common epithelial type was stratified with several layers of squamous epithelium supporting several layers of large cuboidal to columnar cells with pale amphophilic or unstained cytoplasm and cytoplasmic vacuoles, which were strongly alcianophilic (mucinous metaplasia). Mitotic activity or cellular atypia were not apparent. Cystic glands were separated by abundant collagenous stroma. Both the hyperplastic epithelium and the stroma had strong immunoreactivity for progesterone receptor antibody. The pedunculated mass consisted of neoplastic proliferation of intersecting bundles of spindle cells with abundant pale eosinophilic cytoplasm, elongated to plump nuclei and single or multiple nucleoli in scant collagenous stroma. Mitotic figures were rare (2/10 hpf). This tumor was classified as leiomyoma/fibroleiomyoma. By several weeks after hysterectomy, the alopecia had improved dramatically; currently, the guinea pig is clinically normal.

Cystic endometrial hyperplasia is a fairly common condition in bitches as a result of estrogenic 'priming' followed by prolonged progesterone influence. In this guinea pig, histologic findings were more consistent with frequent estrogenic stimulation. Based on the clinical history, uterine changes might have been presented for over two years, which explains the severe (pseudotumoral) proliferation of the endometrium. The prominent mucinous metaplasia of the endometrial epithelium and the keratinizing squamous metaplasia of endometrial glands are distinct from the typical cystic endometrial hyperplasia of bitches. Squamous metaplasia is common in the prostate gland of dogs treated with estrogenic compounds or with estrogen-producing testicular tumors, but is not associated with cystic endometrial hyperplasia of bitches. Concurrent endometrial hyperplasia and leiomyomas (fibroids) have been reported in guinea pigs administered estradiol-like drugs. Endometrial hyperplasia has been studied extensively in the guinea pig as an animal model due to similarities of gestation and the genital tract with humans. This species has also been used to study the pathogenesis of vitamin A deficiency alone or in combination with estrogens due to the close interaction and similar or synergistic effects of both substances on the uterus. Uterine leiomyomas are relatively common in guinea pigs and usually associated with cystic rete ovarii.

# Lymphocystis and Dermal Fibrosarcoma in a New York Walleye

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An adult walleye (*Sander vitreus*) was captured in the spring from Oneida Lake in New York and was found to have multiple discrete skin masses of two different morphologies on the fins and skin. The first morphologic type was characterized by 2 to 4 cm in diameter, multilobulated, glistening, and pink to tan masses. On section, these masses were composed of several small, glistening loculated structures. Histologically, these discrete masses were composed of numerous fibroblasts with marked hypertrophy, consistent with lymphocystis. Lymphocystis is an iridoviral-induced skin disease which is characterized by marked fibroblast hypertrophy and a concurrent mononuclear inflammatory infiltrate. The second morphologic type was characterized by 0.5 to 3 cm in diameter, multilobulated, tan, smooth, and masses which protruded from the surface. These masses were composed of neoplastic spindle cells arranged in streams and interlacing bundles, consistent with a fibrosarcoma. The neoplasm was surrounded by deep aggregates of mononuclear cells. Dermal fibrosarcomas are benign neoplasms seasonally induced by retroviral infection and are often shed at the end of spring. This slide demonstrates two common forms of viral induced skin masses found on a wide variety of freshwater and marine fish.

## Neuronal storage disease in captive aged coatis (*Nasua nasua*)

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From 1983 to 2005, 14 coatis from the collection at the Smithsonian National Zoological Park died. During routine histologic examination of tissues, 5 of 14 demonstrated polysaccharide inclusion bodies in parasympathetic ganglia. Three of these cases were closely related (one mother and two daughters), and all were geriatric (ages 9-17 years). Chronic dermatitis and endocrine gland adenomas were common and occurred rarely in age matched controls. All cases exhibited lethargy and anorexia prior to death. Significant clinical conditions varied and included renal failure, hepatic necrosis, endocrine hepatopathy, metastatic adenocarcinoma, and cystitis with urolithiasis. Euthanasia due to poor prognosis was the manner of death for four of the five. The remaining coati died naturally.

At necropsy, nutritional condition ranged from adequate (3/5) to poor (2/5). Hepatic abnormalities were present in all cases, and primary lesions included moderate multifocal necrosis, endocrine-like hepatopathy, chronic focal hepatitis with focal nodular hyperplasia, telangiectasis, and edema. Only one coati had gross intestinal lesions characterized by multifocal necrotic enteritis; previous routine fecal exams were negative. Central nervous system lesions included one focal mild nonsuppurative encephalitis and one compressive lesion due to carcinoma metastases to bone.

Gross intestinal lesions correlate histologically with expansion of the small intestinal mucosa by a lymphoplasmacytic infiltrate containing Mott cells and eosinophils. The infiltrate often extends adluminally, where a necrotic membrane effaces the villar tips, and less often extends transmurally, where it is associated with thrombi in submucosal vessels. There is a diffuse mild increase in lymphocytes and plasma cells in the colonic mucosa and few, concentrated, lymphoplasmacytic aggregates in the submucosa. One case with no gross intestinal lesions has a mild lymphosuppurative infiltrate in the colonic mucosa and unusual basophilic cytoplasmic inclusion bodies in myocytes of the lamina propria.

Neuronal cytoplasmic inclusions are present in mucosal ganglia of the esophagus, stomach, small intestine, and colon and less commonly in ganglia associated with the mesentery and adrenal glands. There are up to seven inclusions in neurons that have them, appearing as homogenous, eosinophilic, oval structures up to 10- $\mu$ m-diameter, surrounded by an expanded clear cytoplasm. About half of these structures contain tiny basophilic granules.

Special staining and electron microscopy performed by the Armed Forces Institute of Pathology reveal that these inclusions are PAS positive and diastase resistant and are ultrastructurally composed of amorphous to filamentous accumulations of polysaccharide with an electron dense periphery of granular glycogen that is not membrane bound. There is no evidence of viral etiology. There is no history of intestinal dysmotility (i.e. vomiting, diarrhea, abdominal bloating). All cases died of old age-related diseases. These inclusions may be a degenerative change associated with aging and alterations in carbohydrate metabolism. We speculate that there is no apparent clinical significance to this condition, and it is likely genetic.

## Liver Disease in a Cat

*Dodd Sledge, Ralph Common, Matti Kiupel*

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### **Signalment: 4 year old, male castrated Siamese cat**

The provided history indicated that the owner left this cat on a Thursday and returned home Sunday to find the cat extremely lethargic and vomiting. Mucous membranes were pale and his gait was wobbly. On presentation to a veterinarian the cat's temperature was 103°F, but dropped to 96°F within several hours. There was a marked neutrophilia and monocytosis, moderate anemia, and mild thrombocytopenia. ALT was markedly increased. Supportive treatment for shock was initiated, but the cat died within 3-4 minutes.

A 3.6 kg, approximately 4 year old, castrated male Siamese cat was submitted for necropsy. The carcass was in good body condition (4/9 BCS) and autolysis was minimal. The peritoneal cavity contained approximately 120mls of free blood. The liver was moderately enlarged with slightly rounded edges, diffusely pale tan, and had a moderately reticulated appearance. There were multifocal areas of parenchymal hemorrhage that outlined individual lobules and affected approximately 30% of the total liver surface. A large blood clot was focally adhered to the hepatic capsule of the right lateral liver lobe. No other significant lesions were observed grossly.

Histologically, spaces of Disse throughout the liver were expanded by glassy, pale amphophilic, homogenous and amorphous material. There were multifocal areas of necrosis randomly distributed throughout the liver that were often associated with markedly dilated blood-filled sinusoids. The described intrasinusoidal deposits failed to stain with crystal violet, were not birefringent with Congo red staining and examination under polarized light, and did not fluoresce on staining with thioflavine and examination with fluorescence microscopy. Electron microscopy showed deposition of loosely arranged, haphazardly oriented, nonbranching, linear, 8-10nm in diameter fibrils within the space of Disse and within hepatic sinusoids.

### **Diagnosis: Severe hepatic amyloidosis with multifocal hepatic necrosis, telangiectasia and hepatic rupture**

While intrasinusoidal material failed to stain with histochemical techniques generally used to stain for amyloid, electron microscopic examination was most consistent with hepatic amyloidosis. This case demonstrates that while histochemical stains are often useful to distinguish amyloid, they are not perfectly sensitive or specific. Amyloidosis in cats has been well described in Abyssinians and Siamese cats and in both breeds has been associated with a familial pattern of inheritance. The underlying defect leading to atypical deposition of amyloid, however, is unclear. In Abyssinians, amyloid deposits predominately within the medullary interstitium of the kidneys. In Siamese and to a lesser degree other oriental breeds, amyloid predominately accumulates within the liver. In both breeds, the deposits of amyloid have been shown to represent amyloid A (AA). Sequencing of amino acids has shown that while this AA is homologous in both breeds, there is a 2 amino acid difference in Siamese cats, potentially accounting for the difference in deposition site. Hepatic amyloidosis in Siamese cats is often associated with hepatic rupture as observed in this case possibly due the increased friability of the liver.

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## Adult ascariasis in the liver of a pig (*Sus scrofa domestica*)

Molly H. Seavey, Kurt J. Williams

Diagnostic Center for Population and Animal Health, Michigan State University

A three month old female pig was presented for euthanasia and necropsy. Prior to euthanasia, the pig was reported to have dyspnea, cough, diarrhea, and lethargy.

On gross examination, there were multifocal to coalescing pale nodules on the serosal surface of the liver and extending into the parenchyma. These nodules extruded white pasty material on cut section. Within some of these nodules and corresponding ectatic bile ducts, there were worms measuring approximately 20 cm long and 1 mm wide. Within the lungs, there was a severe bronchopneumonia.

Microscopically, within the liver, there were multifocal parasitic granulomas centered over dilated bile ducts. Granulomas were composed of a fibrous periphery and central eosinophilic and suppurative cores. Few ectatic bile ducts contained intraluminal obstructive cross sections of adult nematodes. These nematodes were characterized by a thick cuticle measuring approximately 15 microns. They had coelmyarian musculature, a large digestive tract, and prominent lateral cords. There was no microscopic evidence of parasitic disease in any other organs.

Adults of the large roundworm, *Ascaris suum*, are found principally in the small intestine but may migrate aberrantly into the stomach or bile ducts. Large numbers of eggs are produced (as many as 250,000/day) and they can develop to the infective stage (containing the L<sub>3</sub> larva) in 2-3 weeks in warm conditions. The eggs are resistant to chemical agents, but conditions with low humidity, heat, or direct sunlight may reduce their survival significantly.

After ingestion of the larvated eggs, hatching occurs in the small intestine. Within two hours, numerous small submucosal hemorrhages may be seen in the wall of the duodenum and anterior jejunum as the larvae penetrate to the hepatic portal system. Larvae arrive in the liver where they molt to the L<sub>3</sub>. The L<sub>3</sub> leave the liver and travel to the lungs via the bloodstream. They break out of the capillaries into the alveoli where they molt; the L<sub>4</sub> migrate up the bronchioles, bronchi, and trachea to the pharynx. The L<sub>4</sub> are swallowed and swept to the small intestine where they grow rapidly and molt to the L<sub>5</sub>. Patency occurs at 35-60 days. Longevity is 6 months to over a year.

Clinical debilitation in this case was most likely due to the severe pneumonia caused by *Pasteurella multocida* and *Mycoplasma hyorhinis*. Qualitative fecal revealed many *Balantidium coli* cysts and few *Ascaris suum* eggs. A pig of any age may be exposed to ascarid eggs via feed and water, but most commonly the sources are contaminated floors and dirt lots. Earthworms and dung beetles may serve as reservoirs of infection after ingesting larvated eggs. Pigs may acquire eggs while suckling, when the pasty eggs stick to the mammary glands. There is no evidence of prenatal infection as seen with other ascarid species.

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# Nocardioform Placentitis in a Horse

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A maiden Thoroughbred mare, located in central Kentucky, delivered a live foal approximately 2½ weeks prior to the expected foaling date. The placenta was submitted to the Livestock Disease Diagnostic Center (LDDC) of the University of Kentucky for evaluation.

Examination of the placenta revealed a large, solitary lesion of the chorionic surface. Specifically, there was a moderate amount of opaque, tan to brown, mucoid material over the cranial aspect of the body and extending to the base of the horns. The underlying chorion was pale and associated with a reduced density of villi. Microscopic examination of the affected area revealed a moderately thick surface layer of globular and amorphous, eosinophilic material, aggregates of mixed inflammatory cells, and clusters of branching, filamentous bacteria overlying blunted and necrotic villi. Moderate numbers of neutrophils, lymphocytes, and macrophages were present throughout the remaining villi and superficial chorion.

Aerobic bacterial culture of the affected area of placenta yielded numerous, branching, Gram-positive bacilli, as well as other, nonpathogenic, bacteria. PCR testing of the filamentous bacteria confirmed the presence of both *Crossiella equi* and *Amycolatopsis* sp. Fluorescent antibody testing did not detect antigens of *Leptospira* sp. and no viruses, including no equine herpesvirus, were isolated.

Diagnosis: Chronic-active, locally extensive placentitis due to both *Crossiella equi* and *Amycolatopsis* sp., a.k.a. nocardioform placentitis.

So-called nocardioform placentitis was first described as a cause of placental insufficiency, late term abortion, stillbirth, and the premature delivery of weak foals in horses in central Kentucky in the mid-1980s.<sup>1-5</sup> The organisms associated with these cases have recently been classified as *Crossiella equi*, *Amycolatopsis kentuckyensis*, *Amycolatopsis lexingtonensis*, and *Amycolatopsis pretoriensis*.<sup>6,7</sup> Isolated cases of the same condition have been reported in South Africa<sup>8</sup>, Italy<sup>9</sup>, and Florida.<sup>10</sup>

The LDDC diagnosed 606 cases of nocardioform placentitis during the 15-year period from 1991 to 2005, inclusive, with a low of 2 cases in 1993, a high of 144 cases in 1999, and a mean of 40.4 cases per year, representing approximately 15% of all cases of equine placentitis. This case is the first of those examined at the LDDC in which both types of nocardioform bacteria - *Crossiella equi* and *Amycolatopsis* sp. - were identified since PCR testing became routinely available in 2004.

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10. Christensen BW, Roberts JF, Pozor MA, et al. *J Am Vet Med Assoc* 2006;228:1234-1239.

## **Hypertrophic osteodystrophy and soft tissue mineralization in the Weimaraner**

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A 19 week old Weimaraner puppy presented to the referring veterinarian with a history of lameness, joint swelling, and fever. The clinical signs progressed rapidly to an inability to walk, lethargy, joint pain, and eventual death. The animal was submitted for necropsy examination. Grossly, there was swelling around the stifles and costochondral junctions with periarticular hemorrhage. On section of the long bones, there was a tan, soft linear area along the metaphyseal margin of the epiphyseal growth plate. The cortical bone adjacent to this area was markedly thin. Histologically, sections of long bone exhibited the characteristic changes associated with hypertrophic osteodystrophy (HOD), including marked necrosis with a dense band of neutrophils at the metaphyseal border of the physis, retained cartilage cores, and thinning of the cortical bone. Additionally, there was extensive perivascular mononuclear inflammation and mineralization of the arteries in the sciatic nerves and within the portal and centrilobular areas of the liver. These slides demonstrate the characteristic histology of HOD in the Weimaraner as well as the commonly associated soft tissue mineralization. Soft tissue mineralization associated with HOD will be further discussed in the context of two additional cases of Weimaraner puppies examined at the Oklahoma Animal Disease Diagnostic Laboratory.

## **Focal Symmetrical Poliomyelomalacia Associated with Selenium Toxicosis in Two Grower Pigs**

*Jeffrey R. Hayes*

Ohio Animal Disease Diagnostic Laboratory, Reynoldsburg, Ohio

Four of five recently purchased 4-H show pigs developed acute onset of paresis of the rear limbs or paralysis of all four limbs, which according to the owner, manifested essentially overnight. One pig could get up on its front legs and dragged its rear limbs, and two other hogs were in lateral recumbency and unable to move. All pigs appeared to be alert, were afebrile, had sensation present in the toes, and would eat and drink if feed and water were presented to them. One pen mate and two pigs in an adjacent pen were unaffected. Two live 3-4-month-old barrows, one 58 pound Yorkshire and one 97 pound Duroc-crossbred, were presented for euthanasia and necropsy after a 5 day course of illness. The Duroc-crossbred hog tried unsuccessfully to rise, and had marked bilateral paralysis of the rear limbs. The Yorkshire hog presented in lateral recumbency and could not move, exhibiting minimal withdrawal reflexes to mildly painful stimuli. Both barrows were alert and aware of their surroundings. Neither pig was blind or exhibited seizures, opisthotonus, nystagmus, or hyperesthesia to sound or touch. Differential diagnosis included vertebral fracture (trauma), vertebral or spinal cord abscesses, arsenic and salt toxicosis, pseudorabies, enterovirus and encephalomyocarditis infections. The pigs were fed a premium hog feed obtained from a new supplier for the last 7 days prior to onset of signs.

No significant gross lesions were observed at necropsy other than local hemorrhage and necrosis in muscles of the left shoulder of one pig and in the thigh muscles of the other pig, compatible with injection site injury. Both pigs had dry firm feces (constipation). During processing of the spinal cords, faint depressions could be seen in ventral horns of the cervical and lumbar spinal cord gray matter.

Microscopic lesions were primarily observed in the spinal cord of each hog. Lesions in the larger pig were confined to the cervical and lumbar segments, whereas 4 of eight thoracic cord sections were also affected, in addition to cervical and lumbar sections, in the smaller, more severely affected pig. There is extensive loss of architecture in the ventral horns of the gray matter in a bilateral and symmetrical distribution, characterized by irregular expanses of clear space (malacia) accompanied by marked infiltrates of large round cells with vacuolated eosinophilic cytoplasm (gitter cells) admixed with low to moderate numbers of eosinophils. Multifocally, proliferations of glial cells are noted. Neuronal degeneration and necrosis are demonstrated by loss of Nissl substance (central chromatolysis) and shrunken cytoplasmic margins and hypereosinophilia, respectively. Other changes in the gray matter include endothelial cell hypertrophy of capillaries in affected regions. In the white matter adjacent to affected gray matter, there are swollen myelin sheaths, swollen axon cylinders (spheroids) and loss of axons, primarily in lateral and ventral fasciculi. The morphologic diagnosis was marked, bilaterally symmetrical poliomyelomalacia.

Samples of serum, liver and brain from each pig were submitted to the Diagnostic Center for Population and Animal Health (DCPAH) at Michigan State University, as was a sample of the feed provided to the pigs at the time of onset of signs. Serum selenium levels were 899 and 707 ng/mL

(reference range 130-200ng/mL), and liver selenium levels were 8.62 and 6.73 ug/g (1.0-2.5 ug/g reference range). Sodium levels of fresh brain tissue were 1330 and 1440 ppm (reference range 1850-2030 ppm). The feed selenium level was 93.0 ppm.

Signalment, clinical signs, nature and distribution of microscopic lesions are consistent with a diagnosis of selenium toxicosis, which was supported by elevated serum and liver selenium levels in each pig. The selenium level in the feed sample was markedly elevated, also. Feed levels of selenium greater than 7 ppm are regarded as toxic. Selenium toxicosis should be included in the differential diagnosis for neurologic conditions in swine, warranting consideration of feed selenium levels.

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## Mineralization of the brain stem in a horse with clinical neurological signs

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An 18 year old horse with a clinical history of neurological signs of 72 hrs duration was submitted for post-mortem examination to the San Bernardino branch of the California Animal Health and Food Safety Laboratory of the University of California, Davis. Post mortem examination revealed focal symmetrical areas of mineralization and malacia in the cerebellar white matter in areas corresponding to deep cerebellar nuclei. Histologically, severe mineralization of blood vessels (arteries and veins) and neuropil surrounding these vessels was prominent in these areas. Few neurons and glial cells were degenerated or necrotic in areas surrounding vascular and parenchymal mineralization. Mild to moderate mineralization of blood vessels but not brain parenchyma was also observed in the internal capsule of this horse. No inflammatory infiltrate was observed in any of the areas of mineralization or elsewhere in the central nervous system of the horse. Ancillary tests performed included virus isolation, PCR for West Nile virus and equine herpesvirus-type 1, western blot for *Sarcocystis neurona*, serology for EEE, WEE, VEE and SLE, and determination of brain cholinesterase. Results of all these tests were unrewarding. Mineralization of blood vessels in aged horses is usually considered an incidental finding without clinical significance. However, the severe mineralization observed in the brain of this horse with neurological signs, coupled with negative results for other possible etiologies suggests a relationship between mineralization and the clinical signs observed. The etiology of the lesion remains undetermined.

## **Bovine Herpesvirus-1 Enteritis in a Neonatal Calf**

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Tissues, reportedly from a 2-week-old, Holstein bull calf, were submitted to Purdue University Animal Disease Diagnostic Laboratory for histologic examination, bacteriology, and virology. According to the history provided, 50 of 72 (69%) calves on this farm developed diarrhea with subsequent improvement, then began to decline over the following 24 hours. One animal had died at the time of submission. According to the referring veterinarian, on gross examination, the kidneys appeared swollen; the liver was mottled yellow with multiple, scattered, white foci; and the intestinal wall appeared thin with adherent ingesta on the mucosal surface. Sections of small intestine and liver were examined histologically.

Histologically, the small intestinal mucosa and submucosa contained multiple necrotic foci characterized by loss of tissue architecture; accumulation of eosinophilic cell debris, and pyknotic and karyorrhexic nuclear debris; and infiltration of lymphocytes, histiocytes, few plasma cells, and neutrophils. In areas of necrosis, few cells had margined chromatin and contained large, eosinophilic, intranuclear inclusion bodies. Few mucosal vessels contained fibrin thrombi. In less severely affected areas, intestinal villi were blunt and fused with segmental epithelial degeneration and necrosis. Some intestinal crypts were dilated, filled with degenerate neutrophils and lined by attenuated, low cuboidal epithelium with few individual, scattered, necrotic cells. The lamina propria was expanded by lymphocytes, plasma cells, and few neutrophils. Necrotic cell debris, admixed with bacterial colonies and degenerate neutrophils, were distributed throughout the lumen. The liver contained multiple, scattered necrotic foci characterized by loss of hepatocellular architecture, accumulation of cell debris and karyorrhexic debris, and infiltration of many histiocytes. Occasional cells at margins of necrotic foci contained putative intranuclear inclusion bodies.

Bovine herpesvirus-1 (BHV-1, infectious bovine rhinotracheitis virus) was isolated from the small intestine and BHV-1 antigen was detected in small intestinal necrotic foci by immunohistochemistry. *Escherichia coli* was isolated from the small intestine. Bovine coronavirus and rotavirus were not detected by fluorescent antibody testing.

Bovine herpesvirus-1 infection in neonatal calves can result in multisystemic disease, which often involves the alimentary system. Multiple necrotic foci are most commonly present in the esophagus and forestomachs, although lesions can also be present in the colon and small intestine, as seen in this case. Additional necrotic foci can be seen in other organs including the liver, adrenal gland, and lymphoid tissues. Systemic BHV-1 infections in neonatal calves have been associated with colostrum deprivation.

## Suspected Feed-Associated Renal Toxicity in a Cat

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**Clinical history:** An eleven year old neutered Siamese cat was referred to the Michigan State University Veterinary Teaching Hospital with a reported history of renal failure. The animal was eating a “soft-moist” pouched food which was included in the batches of food recalled by Menu Foods. The renal chemistry abnormalities were as follows: BUN: 182mg/dL (ref: 18-41); Creatinine: 14.8mg/dL (ref: 0.7-2.2); Albumin: 2.2g/dL (ref: 3-4.5)

**Gross lesions:** The kidneys were swollen, diffusely pale tan, and had prominent subcapsular veins, bilaterally. Bilaterally, more than 100 yellow-white crystalline granules peppered the renal interstitium, diffusely; the greatest concentration of these crystals was at level of the papillae and pelves. The renal pelves were moderately dilated. There were hemorrhages on the caudoventral and dorsolateral urinary bladder serosa, which measured 2mm x 4mm and 5mm x 5mm, respectively. There were multifocal petechial hemorrhages on the urinary bladder serosa.

**Histopathologic description:** In the kidney, there was multifocal to locally extensive proximal tubular degeneration, necrosis, and sloughing, with abundant intralésional shattered, circular, refractile green-translucent crystals. There was minimal periarterial edema. There was also a multifocal and coalescing lymphoplasmacytic infiltrate within the renal interstitium, primarily within the cortex.

**Morphologic diagnosis:**

**Kidneys:** Severe, multifocal and locally extensive tubular nephrosis, with intralésional mineral

**Toxicologic analysis:** GC/MS: positive for melamine, cyanuric acid, and ammeline

**Comments:** Melamine (C<sub>3</sub>H<sub>6</sub>N<sub>6</sub>) was first synthesized in 1834. It is used to make a durable, heat-resistant and fire-retardant plastic, which is used in kitchenware, floor tiles, and flame-retardant fabrics, among others. It is created through the endothermic decomposition of urea, and the crystallization of the reactions product, cyanic acid. Further crystallization and washing of melamine for concentrated use creates a waste product, composed of approximately 70% melamine and 20-30% oxytriazines (cyanuric acid, ammeline, and ammeline; all are also metabolites of melamine). It is the solid concentrate of these products, or the parent concentrated melamine itself, which was added to animal feed to increase the protein concentration in the recent Menu Foods recall of pet foods made with wheat gluten imported from China. It is speculated that melamine, cyanuric acid, ammeline, and ammeline have additive effects to create a level of toxicity not present with the individual chemicals. Hydrogen bonding of melamine and cyanuric acid creates a crystalline lattice structure, which is the proposed composition of the crystals found in the kidneys and urine of animals in renal failure which were consuming the recalled foods.

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## **Mycotic dermatitis and microfilaremia in a chameleon**

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This wild-caught male panther chameleon (*Furcifer pardalis*) developed anorexia and lethargy progressing to dehydration and death despite supportive fluid therapy.

Microscopic findings include microfilaremia, most evident in the cardiac section. Subcutaneous granulomatous foci in one of the skin sections are believed to correspond to the location of degenerated adult filarid worms (likely *Foleyella spp.*) a common infection in these reptiles. Intact intracoelomic worms were recovered during necropsy but not submitted for identification. Also evident are foci of hyperkeratosis and epidermal necrosis with intralesional fungal hyphae and bacterial colonies. This debilitated animal was also culture positive for *Salmonella* and had moderate rhinitis, sinusitis and stomatitis (tissues not included).

## **Zygomycotic lymphadenitis in feed lot steers**

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Systemic zygomycosis are caused by different fungi of the class Zygomycetes and affect both humans and animals. The present study was performed in Holstein steers that had been recently slaughtered in one abattoir from Southern California and meat inspection showed tuberculosis-like lesions in the mesenteric lymph nodes. The diagnosis of zygomycosis was based on microscopic lesions, special stains (PAS and Grocott) and DNA sequencing. During 2006, 154 samples were received with tuberculosis-like lesions, from which 141 (92%) were diagnosed as mycotic lymphadenitis. Grossly, the lymph nodes ranged from 1.5 to 48 cm in diameter and were occupied almost entirely by large areas of caseous necrosis with foci of calcification. Histologically the lesions showed a granulomatous pattern, with a necrotic core surrounded by abundant inflammatory infiltrate and fibrosis. Plasma cells were the predominant cell population, followed by lymphocytes, neutrophils and eosinophils. Langhans and foreign body type giant cells were present in all the samples. Numerous fungal hyphae with a bulbous aspect, non-parallel borders, non-dichotomous branching and occasional septae were observed within the necrotic areas and the cytoplasm of the giant cells. Molecular studies revealed the presence of the species *Rhizomucor pusilis* and *Absidia corymbisera*. In conclusion, Zygomycosis should be one of the differential diagnoses to consider in cases of caseous lymphadenitis in cattle.

## Hepatitis associated with *Yersinia pseudotuberculosis* in a Scarlet Macaw

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A four-month old Scarlet Macaw was found dead in an outdoor aviary with no apparent clinical signs. Postmortem examination revealed enlarged liver with numerous 1 to 2 mm white foci scattered through out. Other lesions included a few petechiae on the epicardium, pale spleen and enlarged and congested kidneys.

Histopathology of the liver revealed multifocal necrosis of hepatocytes and infiltration of a mixed population of inflammatory cells randomly scattered through out. There were numerous rod-shaped Gram negative bacteria within the lesions.

**Comments:** *Yersinia pseudotuberculosis* was isolated from the liver, lung and intestine. In addition the bird also had osteomyelitis associated with bacteria. Similar bacteria with out inflammation were seen in the blood vessels of the lung, heart, kidney, intestine, skeletal muscles, etc. Disseminated infection with *Yersinia pseudotuberculosis* is common in psittacines and birds in zoological collection.

## **Atherosclerosis of glomerular capillaries in an adult Yellow Headed Amazon Parrot**

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A Yellow-headed Amazon Parrot was presented for necropsy with clinical findings of poor condition, anorexia, severe exercise intolerance, difficulty in perching and a grade IV/VI sternal heart murmur. Gross postmortem lesions included severely thickened major vessels of the heart and enlarged kidneys with pale foci.

Histopathology of the kidneys revealed prominent glomeruli with enlarged and thickened capillaries associated with pale material. The material in the glomeruli was positive for lipid by Oil red O stain. Many of the cells lining the tubules also contained lipid in their cytoplasm.

**Comments:** The bird also had severe myocardial degeneration and necrosis associated with atherosclerosis of the major vessels and coronary arteries of the heart. In addition severe atherosclerosis was noted in the carotids, thoracic and abdominal aorta and arteries in the liver, lung, mesentery, proventriculus, gizzard, skin, feather follicles, *etc.* Atherosclerosis of the major vessels is very common in psittacines but not of the coronary arteries or other vessels including glomerular capillaries.

## **Abortion, Hydranencephaly and Ocular Defects Attributable to Bluetongue Virus in a Herd of Cattle**

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An increased incidence of stillbirths and birth of dummy calves was noted in a herd of 250 pregnant cattle. Six late term abortions occurred, and there were 8 calves born dead or with neurological signs out of 80 cows that had calved. Abnormal calves were submitted for clinical examination, which revealed horizontal nystagmus, opisthotonus, inability to stand, scleral hemorrhages, muscle fasciculations, chewing movements, bruxism. Clinical illness was not reported in adult cattle. Postmortem examination of 4 of 8 submitted calves revealed severe attenuation and loss of the cerebral cortical tissue that was most severe dorsally but present diffusely. Two calves had cerebellar degeneration (1 grossly) and in one calf, ocular defects characterized by retinal rosettes. Microscopically, the cerebrum had variable neuronal differentiation, with occasional clusters of isolated pyramidal cells. Small dark nuclei, similar to those of periventricular stem cells were interspersed with neurons, sometimes in a laminar pattern. Oligodendrocytes were dispersed among the neurons but had tightly clustered perinuclear processes. There were occasional distended Purkinje cell axons in the cerebellar cortex, but more striking was the mineralization and degeneration in the cerebellar roof nuclei. Similar regions of mineralization were apparent near the hippocampus. Polymerase chain reaction identified bluetongue viral (BTV) sequences in the brains of 4 and 8 calves, but not sequences of BVD or EHD. BTV antibody was identified in the serum of 4 calves (1 presuckle), and in the CSF of 2 calves. One calf had a presuckle serum antibody titer to BVD. Unfortunately, virus could not be isolated successfully to characterize it. Viral isolation is uncommon from animals with fetal anomalies. BTV is not thought by some to be a cause of spontaneous abortion or fetal anomalies in cattle, although it has been reported to cause disease in adult cattle in the Missouri-Arkansas geographical area. It is known that BTV is vasculopathic in both sheep and cattle, but it is uncertain how this relates to the development of hydranencephaly. The maximally susceptible period for developing anomalies in cattle, 130-160 gestational, days coincided with a period of heavy rain in the location of the farm.

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## **Orbital Meningioma with Chondroid and Osseous Metaplasia in a Dog**

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A 3 year old neutered male mixed breed dog was evaluated for exophthalmos of the left eye that had progressed over 2 months and was associated with mild dorsolateral strabismus. There was mild epiphora, nictating membrane protrusion, and increased non-painful resistance to retropulsion of the eye, leading to a diagnosis of retrobulbar mass. The differential diagnosis included nasal, conjunctival and salivary neoplasms, osteosarcoma of the orbit, meningioma, fibrous histiocytoma and salivary mucocele. Ophthalmoscopic examination revealed indentation of the central-medial non-tapedal fundus. Removal of the eye, retrobulbar muscles and mass revealed an infiltrative neoplasm surrounding the optic nerve and occupying much of the retrobulbar fat. The mass consisted of polyhedral to plump fusiform cells arranged in clusters, sheets and bundles, and separated by dense collagen. Although the mass was generally surrounded by fat, individual tumor cells extended incrementally between adipocytes at margins of growth. Within the mass, clusters of tumor cells elaborated blue mucinous matrix, and well defined focal areas of chondrous and osseous differentiation were observed. Immunohistochemical staining demonstrated that cells comprising the mass and those becoming embedded in matrix were positive for cytoplasmic expression of S100 protein and neuron-specific enolase (NSE), as well as for vimentin. The mass did not express GFAP intermediate filament by immunohistochemical testing. Retinal ganglion cells were reduced in number, presumably due to optic nerve compression. A diagnosis of meningioma was made, and the mass occurred near the optic nerve and engulfed the ciliary ganglion. Meningiomas located within the orbit more frequently have the characteristics of mucinous interstitial matrix and focal metaplastic bone and cartilage than do those arising elsewhere, and 3/3 such tumors exhibited these characteristics in a recent case series.

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# Multisystemic Chlorellosis Involving the Tongue and Spinal Meninges of a Dog

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A 9 year old neutered female Golden retriever dog was presented for halitosis, weight loss, and progressive difficulty walking for a week period prior to presentation. Inability to urinate had been noted for the last two days. Similar ataxia had developed 3.5 years previously. A clinical diagnosis of coccidiomycosis was made at that time and the dog was treated with fluconazole, with improvement. At presentation, the dog needed help to rise but could walk with a crouching gait. Conscious proprioception was reduced, but the tail was flaccid, the urinary bladder enlarged and expressible and anal tone was absent. Whereas painful responses could not be elicited by stimulating the anus or tail, lumbar pain was increased. During anesthesia induction for imaging, an irregular green-brown mass was noted on the surface of the tongue that extended into adjacent muscle. MR of the spine revealed an enhancing extradural mass lesion, predominantly on the left side, between L4 and L6, which also extended into adjacent muscle. Biopsy findings from the tongue were consistent with protothecosis, and the patient was euthanized. Grossly, sublumbar and mesenteric lymph nodes were enlarged and green. Green discoloration of the liver was also present, in addition to the mass in the tongue. The muscle around the spinal cord contained a green infiltrate, which was associated with similar extradural material in the vertebral canal. Microscopically, the lesions were characterized by modest granulomatous to pyogranulomatous inflammation. Most of the spinal involvement was extradural, but small numbers of organisms could also be found within the leptomeninges. Organisms were plentiful and were characterized by a few to many endospores visible within sporangia. GMS and PAS stains highlighted numerous small cytoplasmic granules, which are characteristic of *Chlorella*. The characteristics of the organism identify it as being of algal character, and the pronounced green discoloration of lesions identified it as being *Chlorella* rather than a *Prototheca* species. The spinal cord and nerves were affected by Wallerian degeneration. *Chlorella* infections are much less common than *Prototheca*, and only one case of localized infection has been reported in human being, compared to over 100 cases of protothecosis. Four cases of generalized protothecosis involving the central nervous system have been reported in dogs. Most reports of *Chlorella* infection involve a single animal but multiple animals have been reported with enteric infections in sheep grazing wet pasture.

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