CASE I: 09189WFUHS (AFIP 3165085).

Signalment: 24-year-old, female, cynomolgus macaque (*Macaca fascicularis*).

History: The animal presented with a suspected tooth root abscess in October 2009. Pre-operative blood work was within normal limits and the animal was taken to surgery for extraction of the first right maxillary pre-molar and the second and third molar. This was followed with a post-operative course of clindamycin. Swelling and bleeding persisted in the oral cavity and over the right side of the nose and face after surgery and the extraction sites did not heal. Over the next month bleeding from the oral and right nasal cavity increased. A radiograph revealed osteolysis of the right maxilla and a soft tissue opacity at that site. Euthanasia was elected November 2009.

Of significance in the medical history is an episode of endometriosis in 2007 for which an ovariectomy was performed.

Gross Pathology: The animal was in good body condition and weighed 4.14 kg. Fat stores, muscle mass and hydration status were all adequate. Marked dental tartar was present on all remaining teeth, but most molars had been extracted. A 0.5 cm x 4 cm long oronasal fistula created a communication between the right nasal cavity and the mouth at the site of the most recent surgical extractions. Upon reflection of the facial skin, a 2.5 cm diameter mass of soft grey tissue was present in the right maxillary sinus and the bone of the overlying maxilla was eroded away.

Laboratory Results: CBC and biochemistry panel were within normal limits.

Histopathologic Description: Oronasal tissue: The mass is composed of a dense population of polyhedral to oval neoplastic cells that form nests, packets and cords separated by trabeculae of dense fibrovascular connective tissue. Frequently, within these nests and packets, the cells form small acinar and ductular structures. The cells are approximately 15-20 microns in diameter and contain oval, centrally placed nuclei with dispersed chromatin and rarely apparent nucleoli. Anisocytosis and anisokaryosis are mild and mitotic figures are rare. The mass is well demarcated and unencapsulated with an invasive border from which tumor cells infiltrate irregularly into the surrounding soft tissue, bone and around blood vessels. Neutrophils and lymphocytes are present at the tumor border. Multifocal aggregates of deeply eosinophilic, polyhedral cells with indistinct borders containing small round nuclei (squamous differentiation) punctuate areas of the mass. In areas where the mass infiltrates bone, there is effacement of the alveolar bone (where the tooth roots of the extracted teeth would have rested). Tumor cells replace all cells in the marrow cavities and invade bone of the maxilla. Focal aggregates of osteoblasts and small groups of 3-5 osteoclasts are noted.

By immunohistochemistry the cell cytoplasm stains positive for pancytokeratin in some areas, particularly the glandular and ductular structures; they are negative for vimentin.

Contributor’s Morphologic Diagnosis: Nasal adenocarcinoma with squamous differentiation, right nasal cavity and maxillary sinus.
**Contributor’s Comment:** While periodontal disease was present in this animal, the cause for the suspected tooth root abscess was actually an invasive growth of a nasal adenocarcinoma into the maxilla that also invaded and replaced the alveolar bone and tooth roots on the right upper arcade.

Nasal adenocarcinoma is an uncommonly reported tumor of the upper respiratory tract in non-human primates. There are few reports of nasal adenocarcinomas or carcinomas occurring in non-human primates in the literature.\(^1,7,9\) In humans, it has been associated with various occupational exposures to inhaled substance such as the fine particulate matter found in the woodworking and textiles industries and fumes and vapors common to the chemical industry.\(^6,10\) Experimental work in macaques has also shown that ozone may act as a toxic inducing agent when in contact with the nasal mucosal epithelium.\(^5\) Adenocarcinomas are characteristically composed of glandular structures that usually contain some degree of secretory product. The most common forms are tubular, tubulopapillary and acinar. Mixed patterns are frequent. Low-grade tumors have glandular spaces or papillary fronds lined by cuboidal to columnar cells in a single layer with a round to oval nucleus and inconspicuous nucleoli, whereas high-grade tumors have irregular glandular spaces, more solid sheets of cells and a high mitotic rate with cellular pleomorphism and nuclear atypia. Mucus is the most common secretion in adenocarcinomas and often there is retention, creating cystic spaces. In addition to tubular, tubulopapillary and acinar classifications, adenocarcinomas may be further classed as mucinous or adenocarcinomas with marked desmoplasia (fibrous response). Adenocarcinoma with squamous metaplasia or differentiation is reserved to describe tumors with minor portions containing regular squamous differentiation, as in this case. Adenosquamous carcinoma refers to tumors that are typically highly invasive and have prominent intermixing of adenocarcinomatous and malignant squamous cell components.\(^11\)

**AFIP Diagnosis:** Sinonasal tissue: Adenocarcinoma, salivary gland-type, with extensive squamous differentiation.

**Conference Comment:** Most conference participants favored a diagnosis of adenosquamous carcinoma, primarily mucoepidermoid type. This generated discussion of a differential diagnosis list for this lesion which, in addition to the contributor’s diagnosis of adenocarcinoma with squamous differentiation, would also include adenosquamous carcinoma, undifferentiated carcinoma and carcinosarcoma. Adenosquamous carcinomas are characterized by a mixture of squamous cell carcinoma (SCC) and adenocarcinoma with frequent intracellular carminophilic mucin.\(^2\) Adenosquamous carcinomas of the lung are histologically identical to those in the sinonasal region.\(^2\) Undifferentiated sarcomas of the head and neck are composed of a population of undifferentiated, uniformly chromatic cells with a prominent lymphoplasmacytic infiltrate and absence of squamous or glandular differentiation.\(^2\) Carcinosarcomas have a dominant spindle cell population, sarcoma-like stroma and a minor component of SCC or in situ carcinoma; the neoplastic spindle cells can produce collagen, osteoid and cartilaginous matrix.\(^2\)

This case was also studied in consultation with the AFIP’s Department of Oral and Maxillofacial Pathology, whose pathologists agreed with the contributor’s diagnosis of a sinonasal adenocarcinoma; they further classified the tumor as salivary gland-type. In addition to the extensive squamous differentiation, the subspecialty pathologists also commented on the presence of a prominent myoepithelial component as indicated by immunopositivity for smooth muscle actin.

In humans, glandular malignant neoplasms of the sinonasal tract are classified as salivary and non-salivary types; non-salivary sinonasal adenocarcinoma (ACA) is further subdivided into intestinal-type and non-intestinal-types as outlined below.\(^3,4\)

1. **Non-salivary type**
   a. Intestinal-type adenocarcinoma
      i. Papillary-type
      ii. Colonic-type
      iii. Solid-type
      iv. Mucinous-type
      v. Mixed
   b. Non-intestinal-type
      i. Low-grade
      ii. High-grade
2. **Salivary gland-type**
   a. Adenoid cystic carcinoma
   b. Acinic cell carcinoma
c. Mucoepidermoid carcinoma
d. Epithelial-myoepithelial carcinoma
e. Clear cell carcinoma
f. Other (rarely reported)
   i. Malignant myoepithelioma
   ii. Carcinoma ex pleomorphic adenoma
   iii. Polymorphous low-grade adenocarcinoma
   iv. High grade adenocarcinoma

The intestinal type is histologically similar to intestinal ACA; well-differentiated sinonasal intestinal-type ACAs can have Paneth cells, enteroendocrine cells, and goblet cells and may form villi with a muscularis mucosae.\(^2,4\) Non-intestinal-type sinonasal ACAs are classified as low-grade or high-grade. The histologic appearance can vary between and within tumors, with papillary, oncocytic and clear cell patterns; the high-grade form tends to be more solidly cellular.\(^2,4\)

Salivary gland-type ACAs of the sinonasal tract account for between 5-10% of all sinonasal ACAs in humans.\(^8\) Though histologically similar to salivary gland ACAs, they are a distinct entity in that they arise from seromucous glands and surface epithelium of the nasal cavity and paranasal sinuses.\(^8\) The salivary gland-type tumors are composed of glandular epithelium surrounded by myoepithelial cells.\(^2\) Neoplastic epithelial cells frequently contain intracellular and extracellular mucin which stains with mucicarmine and Alcian blue.\(^8\) Mucin frequently accumulates in small cystic spaces. In contrast to many glandular neoplasms in which neoplastic epithelial cells undergo squamous or ductal differentiation, the squamous component of salivary gland-type ACA arises from myoepithelial cells.\(^8\)

This type of neoplasia in humans can initially present as swelling of the palate and face and loosening of the teeth, which were the presenting clinical signs in this case.\(^3\) These tumors often invade adjacent bone, causing osteolysis. Though considered a malignant neoplasm, these tumors rarely metastasize, though most patients succumb to the effects of locally aggressive invasion.\(^3\)

We would like to thank the Department of Oral and Maxillofacial Pathology for their review of this case, and specifically CAPT Robert Foss, DDS, for his enlightening comments.

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References:
CASE II: YN08-445 (AFIP 3166455).

Signalment: 13-year, 7-month-old, male, sooty mangabey (Cercocebus atys).

History: This adult male sooty mangabey was born at the Field Station of the Yerkes National Primate Research Center. He was diagnosed as being diabetic. Severe preputial edema was noted and due to poor prognosis, he was sacrificed two days later.

Gross Pathology: The animal weighed 13.36 kilograms at necropsy. There was a large amount of turbid, milky white fluid intermixed with adhesions between the gastrointestinal serosa and the mesentery in the abdominal cavity. Multiple, white, firm, ovoid, well-encapsulated structures measuring approximately 0.25-1.5 cm in diameter were present on the diaphragm, peritoneum, mesentery and serosa of the urinary bladder. The mesenteric lymph nodes were enlarged, pale white and firm on sectioning. The prepuce had severe subcutaneous edema.

Laboratory Results: Diabetic. Serology: SIV positive and HTLV1 negative. Microbiology: no significant pathogens were isolated from blood, liver or colon.

Histopathologic Description: Lymph node: The nodules in the abdominal cavity are well-circumscribed by fibroblasts. Severe extensive aggregates of neutrophils, multinucleate giant cells and few eosinophils intermixed with asceptate, branching, fungal hyphae (consistent with a zygomycotic agent) are present at the center of these nodules. The enlarged mesenteric lymph nodes have severe pyogranulomatous infiltrates intermixed with intralesional fungal organisms. Staining with Gomori methenamine silver stain confirmed the presence of a zygomycete.

Contributor’s Morphologic Diagnosis: Severe multifocal granulomatous peritonitis with intralesional fungi (zygomycete).

Contributor’s Comment: Zygomycosis is a relatively new term that refers to a group of uncommon but frequently fatal mycoses caused by fungi of the class Zygomycetes. This mycosis is characterized by subcutaneous, systemic, or rhinocerebral infections. The common diagnostic feature of zygomycetes is that the organisms form infrequently septate hyphae which are significantly broader than other fungi with filamentous tissue forms, e.g. Aspergillus. The hyphae are unpigmented and range from 6-25 µm in diameter. The class Zygomycetes has two orders: Mucorales (genera: Rhizopus, Mucor, Absidia, Mortierella, Rhizomucor among others), members of which cause mucormycosis; and Entomophthorales (genera: Basidiobolus and Conidiobolus), which cause entomophthoromycosis. Zygomycetes are widespread in nature, occurring as soil saprophytes, components of normal skin and hair flora or as common laboratory contaminants. They may enter the host through cutaneous, gastrointestinal or respiratory routes.

The members of the order Mucorales cause most human disease, characterized by a rapidly evolving course, disseminated disease, tissue destruction, and invasion of blood vessels. This disease is associated with preexisting conditions which lead to immunosuppression, such as diabetes, nutritional deficiencies, severe burns, hematologic or oncologic diseases, transplant recipients, and high-risk neonates. Zygomycosis is a rare disease of animals, including dogs, cats, horses, llamas, sheep, pigs and several nonhuman primate species. Both Old World and New World primates are susceptible to the infection, which is rare in prosimians. Ulceration and necrosis of the alimentary tract mucosa are the most frequently described gross lesions.

Culture or immunofluorescence studies, or both, are necessary for species-specific identification of the fungi. In the current case, samples suitable for culture or molecular identification were unavailable. The organisms can be more easily seen and better characterized when stained with periodic acid–Schiff (PAS) or Gomori methenamine silver (GMS) techniques. GMS revealed the presence of aseptate hyphae with broad, irregular, branching often at perpendicular angles (consistent with a Zygomycte) in these granulomas. A negative acid fast bacilli stain precluded the presence of Mycobacterium in these lesions.
**AFIP Diagnosis:** Mesentery: Peritonitis and steatitis, pyogranulomatous and eosinophilic, focally extensive, severe, with fibrosis, many multinucleate macrophage giant cells, and few aseptate fungal hyphae, etiology consistent with zygomycetes.

**Conference Comment:** Conference participants agreed that the etiologic agent was most likely a zygomycete; few ventured beyond that in an attempt to further classify it as a member of either the Mucorales or the Entomophthorales family.

This case was also studied in consultation with the AFIP’s Department of Infectious Disease Pathology, whose subspecialty pathologists favored a member of the Entomophthorales as the etiologic agent because of the marked eosinophilic infiltrate. Infection with Entomophthorales is histologically characterized by multiple pyogranulomas, marked eosinophilic infiltration, foreign body giant cells, and rare fungal hyphae.1 Though not evident in this specimen and, in contrast to Mucorales, Splendore-Hoeplli material often surrounds fungal hyphae, forming an “eosinophilic sleeve” highlighting the hyphae.3 Members of Entomophthorales may exhibit lateral and deep spreading; invasion of the blood vessels is uncommon.1

Entomophthorales is a cause of cutaneous zygomycosis in the horse and, rarely, the dog and cat. Dogs and cats are typically infected with Conidiobolus spp. Gross lesions in the dog consist of poorly circumscribed dermal nodules which spread to form “satellite nodules” followed by necrosis, ulceration, and fistulation with purulent or serosanguineous exudate.3

Horses are most commonly infected with Basidiobolus haaptosporus and Conidiobolus coronatus, which produce round, ulcerated, and usually solitary swellings most often involving the chest, trunk, head and neck. The lesions are pruritic and contain gritty, yellow-white granules referred to as “kunkers” or “leeches”. On cut surface, the granulomas appear as a wavy swath of yellow-white material sharply demarcating the granulomas from the superficial hemorrhage and edema.1

Infection of sheep with C. incongruus produces rhinitis with asymmetrical facial swelling. Infected animals typically die within 7-10 days after showing clinical signs.1

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**References:**

**CASE III:** CSUGP131 (AFIP 3166606).

**Signalment:** 12-week-old, intact female, Dunkin-Hartley, guinea pig (Cavia porcellus).

**History:** This guinea pig was infected with 500 colony forming units (CFU) of Burkholderia pseudomallei by intranasal inoculation. The animal was euthanized 12 days post-inoculation because of deteriorating clinical condition, including pyrexia, lethargy and dyspnea.

**Gross Pathology:** Disseminated throughout the parenchyma of all lung lobes and present rarely within the liver and spleen parenchyma are multifocal, discrete, raised white nodules that measure 1 – 3 mm in diameter. On cut surface,
larger nodules are surrounded by a circumferential pale tan rim of tissue with centralized, white to tan, friable debris (abscesses).

**Histopathologic Description:** Lung: Multifocal, discrete, and often coalescing inflammatory foci effacing approximately 80% of the pulmonary parenchyma are often centered on bronchi and bronchioles and are composed of centralized accumulations of degenerate heterophils surrounded by a periphery of epithelioid histiocytes and admixed fibroblasts. Larger inflammatory foci have abundant central necrosis. The lumen of airways is often filled with degenerate heterophils that multifocally infiltrate through and obscure the bronchiolar and bronchial wall. There is segmental loss and effacement of airway epithelium. Adjacent alveolar septa are expanded up to two times normal by fibrin, edema, heterophils and other mononuclear inflammatory cells. Alveolar spaces contain abundant eosinophilic edema fluid and fibrin strands admixed with alveolar macrophages with foamy cytoplasm. Multifocally, there are rows of enlarged cuboidal pneumocytes lining alveolar septa containing large nuclei with open chromatin (type 2 pneumocyte hyperplasia). Multifocally, peribronchiolar and perivascular connective tissue is expanded by clear edema fluid. Perivascular inflammatory infiltrates are present consisting of macrophages, lymphocytes and heterophils that multifocally infiltrate and obscure the vessel wall. Associated with the inflammation, vessel walls have segmental hyalinization (fibrinoid necrosis) or expansion of the adventitia and segmental obliteration of the wall by fibrous connective tissue that occasionally extends to fill the vascular lumen (not present in all slides). Multifocally, inflammatory infiltrates extend into the pleural connective tissue and are admixed with fibroblasts and mucinous matrix.

**Contributor’s Morphologic Diagnosis:** Lung: Multifocal and coalescing, necrotizing heterophilic and histiocytic bronchopneumonia with fibrosis, pleuritis, and leukocytoclastic to fibrosing and obliterative vasculitis, chronic, severe.

**Etiology:** Burkholderia pseudomallei

**Contributor’s Comment:** The Gram-negative, aerobic, motile bacillus *Burkholderia pseudomallei* is the causative agent of melioidosis and is endemic to the tropical climates of Southeast Asia and Northern Australia. The bacterium is recognized as an important emerging pathogen, accounting for a significant proportion of septicemic mortality in endemic areas and is classified as a select agent. *Burkholderia pseudomallei* is a saprophytic, facultative intracellular bacterium with primary reservoirs being rice paddies, stagnant water, and moist tropical soils. *Burkholderia pseudomallei* can persist for long periods of time, spanning up to 10 years, in low-nutrient conditions, but is not a spore-forming bacterium. Additionally, the bacterium is resilient in adverse conditions including exposure to detergents, acidic pH, and dehydration. There is a strong association between occurrence of melioidosis cases in humans and monsoonal rain season, presumably due to dislodging of the organism from its environmental niche and aerosolization. *Burkholderia pseudomallei* is capable of infecting and surviving within *Acanthamoeba* trophozoites, and it is presumed that factors allowing for intra/protozoal survival are reflected as virulence factors upon invasion of human and animal macrophages.

*Burkholderia pseudomallei* has an extremely wide host range and is a recognized pathogen of human and animal species, most commonly infecting sheep, goats and pigs. However, sporadic cases and small epizootics have been reported in a variety of animal species including non-human primates, kangaroo, wallaby, deer, buffalo, cow, camel, llama, zebra, koala, dog, cat, horse, mule, parrot, rat, hamster, ground squirrels, seal, dolphin and crocodile. Cases in boids, birds and reptiles are rare, and these species are considered to be relatively resistant to *B. pseudomallei*. A wide spectrum of clinical manifestations may arise from infection with *B. pseudomallei* ranging from acute, rapidly progressive, and often fatal septicemia to chronic multi-organ disease with abscess formation. Accepted routes of transmission are via either percutaneous inoculation or inhalation. Disease manifestations in animals include pneumonia, arthritis, orchitis, mastitis, abortion, meningencephalitis, dermatitis, and lymphangitis. Acute disease may manifest in the majority of affected species, and most often occurs in younger animals. Chronic manifestations of the disease appear to be more common. Pneumonia is the most common manifestation of disease in humans and is present in approximately 50% of cases. Differences in human clinical manifestations depending on geographic location are recognized, with a high incidence of prostatic abscesses occurring in Australian males, while up to 40% of Thai children present with suppurative parotitis. Skin and soft-tissue infections are also common manifestations and may be the source for hematogenous spread of the organism. *Burkholderia pseudomallei* is intrinsically resistant to multiple groups of antibiotics, including 3rd generation cephalosporins, penicillins, aminoglycosides, quinolones and macrolides. This limits therapeutic options and accounts for the high rate of therapeutic failure. Ceftazidime and amoxicillin-clavulanate are the current antibiotics of choice in treatment of melioidosis.
The pathogenesis and virulence factors of *B. pseudomallei* are heavily researched and are well described in a recent review. Initial infection occurs via attachment to epithelial cells at the site of inoculation by an unknown molecule presumably associated with the exterior polysaccharide capsule of the bacterium. The organism is capable of invading non-phagocytic and phagocytic cell types. *Burkholderia pseudomallei* employs a type 3 secretion system (T3SS), a system seen in many other virulent bacteria which utilizes effector molecules to manipulate host cell function. Actin cytoskeletal rearrangement in the host cell induced by T3SS may account for cellular invasion. Subsequently, the bacterium is able to escape the initial phagocytic vacuole by way of T3SS effectors and replicate in the cytoplasm. *Burkholderia pseudomallei* can also modulate production of reactive oxygen intermediates in professional phagocytes as an important mechanism of pathogenesis and evade intracellular killing mechanisms. Upon reaching a critical threshold of replication based on quorum sensing, the organism lyses the cell by inducing caspase 1-dependent lysis or apoptosis. This allows for secondary hematogenous dissemination.

The histopathologic features of human melioidosis are described as acute, necrotizing to chronic neutrophilic and granulomatous inflammation. Multinucleated giant cells are a common feature, and in some cases intracellular bacteria within macrophages are so numerous as to resemble globi, a term describing aggregates of bacteria in lepromatous leprosy. Small abscesses may be identified in numerous organs including lung, liver and spleen. Bacterial organisms may be more indistinct with chronic manifestations of melioidosis, as indicated in the provided slides. However, more acute manifestations of the disease in this infection model did display large numbers of intrahistiocytic and extracellular bacilli. Animal models of melioidosis have been developed in several species and may be further expanded given the wide host range of *B. pseudomallei*. However, significant differences in the pathogenesis of disease are recognized in different models and it is thus difficult to assess which model best reflects human disease.

### AFIP Diagnosis:
Lung: Bronchopneumonia, suppurative and necrotizing, diffuse, marked, with alveolar edema, pleuritis, multifocal fibrosis, and rare vasculitis.

### Conference Comment:
Conference participants agreed on a diagnosis of necrotizing and suppurative bronchopneumonia. The etiologic differential diagnosis considered by participants included adenovirus, *Streptococcus zooepidemicus*, *Streptococcus pneumoniae*, *Klebsiella pneumoniae*, and *Bordetella bronchiseptica*. Gross findings in adenoviral pneumonia of guinea pigs typically include consolidation of the cranial and hilar lung lobes; histologic findings include non-suppurative, necrotizing bronchitis and bronchiolitis. Nuclei of infected cells frequently contain characteristic large, basophilic intranuclear viral inclusion bodies.

Of the bacterial etiologies considered by participants, *K. pneumonia* and *B. bronchiseptica* are Gram-negative; histologic findings are typically necrotizing and suppurative owing to the lipopolysaccharide of their cell walls, which damages the endothelium and is a potent activator of leukocytes. The streptococcal organisms usually elicit a fibrinopurulent pneumonia, pleuritis and serositis. The strain of *S. pneumoniae* most often infecting guinea pigs is capsular polysaccharide type 19, though type 4 is occasionally isolated.

Repair of the airways and lungs frequently involves local stem cells. Within the trachea, Clara-like cells and cells within the submucosal glands appear to act as stem cells; basal cells of the trachea and bronchi also appear to have stem cell properties, allowing them to replace injured epithelium. Bronchiolar Clara cells have bipotential properties, as they can differentiate into ciliated epithelial cells or produce additional Clara cells. Bone marrow-derived stem cells may also play a role in pulmonary repair through differentiation into epithelial and mesenchymal cells.

For a review of the primary defense mechanisms of the lung, the reader is encouraged to review Conference 15, Case 2, 2009-2010.

### Contributor:
Colorado State University, College of Veterinary Medicine and Biomedical Sciences

### References:


CASE IV: YALE 174-CASE-1 (AFIP 3169856).

Signalment: 6-8-week-old, female, C3H/HeJ, mouse (Mus musculus).

History: There were unexpected deaths in mice used for feeding Borrelia burgdorferi-infected ticks.

Gross Pathology: At necropsy there was marked bilateral submandibular lymphadenopathy, mild splenomegaly, and marked paucity of abdominal adipose tissue.

Laboratory Results: Serology was negative for corona virus, EDIM, LCMV, Ectromelia, MPV, Sendai virus, TMEV, PVM, and M. pulmonis. Warthin-Starry stain of brain tissue was negative, and cardiac tissue was positive by Warthin-Starry for spirochetes.

Histopathologic Description: Heart: There is a mild to moderate focally extensive inflammatory infiltrate admixed with fibroblast proliferation and necrotic cellular debris centered on the connective tissue surrounding the great vessels at the base of the heart. The inflammatory infiltrate extends multifocally into the wall of the aorta and multifocally into the superficial surrounding myocardium. The inflammatory infiltrate consists of a mixture of macrophages and neutrophils with fewer numbers of lymphocytes and plasma cells. Infrequent spirochetes are observed on Warthin-Starry-stained sections of heart.

Contributor’s Morphologic Diagnosis: Carditis, myocarditis, vasculitis, mild to moderate, chronic active, focally extensive with intralesional spirochete bacteria consistent with Borrelia burgdorferi.

Contributor’s Comment: Lyme disease is a globally occurring, usually nonfatal multisystemic disorder caused by the gram-negative spirochete Borrelia burgdorferi. In the United States, it is the most commonly reported vector-borne disease, most often transmitted by Borrelia-infected nymphal ticks of the genus Ixodes. Lyme disease has been reported in 49 states and the District of Columbia. Ninety-two percent of the cases occurred in 10 states, mainly in the northeast. Elsewhere in the world, infection has occurred in Russia, China, Japan, and Europe. Borrelia is capable of infecting both humans and animals, including mice, rats, hamsters, rabbits, dogs, cattle, and horses. In humans, Lyme disease is characterized by a target-shaped skin rash, neurological symptoms, and various inflammatory processes including arthritis and carditis. Within 2-3 weeks of infection, clinical manifestations in the joints and heart peak, then begin to regress. While 60% of patients in the United States develop arthritis, an estimated 4 to 10% with untreated infections develop cardiac manifestations of the disease. Arthritis often recurs, but carditis is transient and usually present for a few days to several weeks. Common clinical complaints in patients with Lyme carditis include light-headedness, syncope, dyspnea, and heart palpitations. Electrocardiographic abnormalities, such as prolonged P-R intervals and heart block, are often identified, though some patients are asymptomatic. In patients with EKG abnormalities, alternating tachycardia and bradycardia are not uncommon.

Several laboratory animal species are susceptible to experimental Lyme disease. Because of the relative ease by which their genome can be manipulated, mice are currently the most valuable animal model for studying Borrelia infection. Severity of disease varies by strain, with C57BL/6, SJL, and BALB/c being most resistant, and C3H/He being most severely affected. Investigations utilizing transgenic and knockout mice have proven useful for studying the immune system response and pathology associated with B. burgdorferi infection.
In the rodent model, carditis and arthritis are consistent pathologic findings. Neologic disease and skin lesions, though common in humans, are uncommon manifestations of borreliosis in animal models. Histopathologic cardiovascular lesions in human and animal patients with Lyme disease may occur in any layer of the heart. In some cases, *B. burgdorferi* spirochetes may be demonstrated in cardiac tissue by indirect immunofluorescence or silver staining. Organisms typically reside in the connective tissue at the AV junction, epicardium and, occasionally, in the myocardium. In addition, vasculitis in small and large intramyocardial vessels is observed. Macrophages tend to be the predominant inflammatory cell type, evident by immunohistochemistry at 7 days post-infection, followed by plasma cells and neutrophils. This is in contrast to joint lesions, in which neutrophils are the predominant inflammatory cell type. Poliarthritis after infection with *B. burgdorferi* occurs in mice of all strains, but the severity, frequency, and extent of arthritis is strain and age-dependent. Swelling of the joints, especially the tibiotarsal joints, is often noted.

In the case presented, 12 C3H/HeJ mice were used as feeders for *Ixodes scapularis* nymphal ticks infected with *B. burgdorferi*. The clone of *Borrelia* usually used is cN40, a low passage isolate with proven infectivity for laboratory mice. Eight nymphs are placed on the mouse, allowed to feed for 72 hours, then removed. This procedure is not normally associated with mortality in the feeder mice. Cardiac lesions, with or without joint lesions, are common with this clone. Brain lesions are not typically observed. However, in this case, a new batch of ticks infected with *B. burgdorferi* 206 was used and 8 of 12 mice were found dead between days 11 and 13 post-feeding. The ticks tested negative for other pathogenic agents, including *Ehrlichia*, *Babesia*, and *Anaplasma*. One mouse was submitted for necropsy to rule out other infectious causes for the acute deaths observed in these mice. The lack of any other significant pathologic lesions and the presence of the lesions noted within the brain and heart, along with the presence of spirochetes in the heart by Warthin-Starry stain, suggest the mice died secondary to cardiac complications of *B. burgdorferi*. The investigator euthanized the remainder of the mice and eliminated all the ticks from this batch. The HE stained slides submitted for this conference came from mice experimentally infected with *B. burgdorferi* from other studies, where all have the classic carditis seen in this experimental model and were known positive for *B. burgdorferi*.

**AFIP Diagnosis:** Heart: Endomyocarditis, valvular and atrial, lymphohistiocytic and neutrophilic, multifocal, mild to moderate, with focal epicarditis.

**Conference Comment:** The contributor provides an excellent overview of borreliosis in laboratory rodents.

Conference participants commented on slide variability; some sections had lymph nodes that exhibited diffuse hyperplasia. Additionally, focal epicarditis was present in some sections.

Participants reviewed various non-viral tick-borne diseases of veterinary importance with respect to the pathogen and its associated vector. The chart below provides a brief summary.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Tick Vector</th>
<th>Susceptible host(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ehrlichia canis</em></td>
<td><em>Rhipicephalus sanguineus,</em></td>
<td>Dog, ± cat</td>
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<td></td>
<td><em>Dermacentor variabilis</em></td>
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<tr>
<td><em>Ehrlichia ruminantium</em></td>
<td><em>Amblyomma hebraeum</em></td>
<td>Cow, sheep, goat, dog</td>
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<tr>
<td><em>Ehrlichia equi</em></td>
<td><em>Ixodes pacificus</em></td>
<td>Horse, dog, man</td>
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<td><em>Dermacentor spp.</em></td>
<td>Cow</td>
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<td><em>Anaplasma phagocytophilum</em></td>
<td><em>Ixodes scapularis</em></td>
<td>Dog, cat, horse, man</td>
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<td><em>Dermacentor spp.</em>, <em>Rhipicephalus</em></td>
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<td><em>spp.</em>, <em>Amblyomma spp.</em></td>
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<td><em>Francisella tularensis</em></td>
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<td>Many</td>
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<td><em>Amblyomma americanum</em></td>
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<td><em>Babesia canis canis</em></td>
<td><em>Dermacentor reticulatus</em></td>
<td>Dog</td>
</tr>
<tr>
<td><em>Babesia gibsoni</em></td>
<td><em>Rhipicephalus sanguineus</em></td>
<td>Dog</td>
</tr>
</tbody>
</table>
Lyme borreliosis in the dog can clinically manifest as a systemic illness, arthritis, renal disease and meningitis. Polyarthritis and shifting leg lameness are common manifestations, with the limbs closest to the tick attachment site affected first. Apparent resolution of the lameness in untreated dogs does not necessarily equate to a halt in joint pathology, as progressive, non-erosive arthritis is commonly detected. Renal disease in infected dogs is clinically evident as azotemia, uremia, proteinuria, peripheral edema and body cavity effusions; these findings are consistent with acute renal failure. Meningitis may manifest later in the disease process in humans, and lesions have been experimentally reproduced in dogs though clinical signs were not evident.

For a review of the pathogenesis and microscopic changes in the kidney, the reader is encouraged to review WSC 2009-2010, Conference 25, Case 4.

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**References:**