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Wyeth Research
641 Ridge Road
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CASE I – 03-10192 (AFIP 2942336)

Signalment: 7-year-old Polypay ewe.

History: This ewe was a chronic poor doer and was positive serologically for ovine progressive pneumonia virus.

Gross Pathology: The ewe was in poor body condition. The lungs were heavy and did not collapse. The cranial ventral portions were consolidated and contained several abscesses. Hilar lymph nodes and gastrohepatic lymph nodes were enlarged and replaced by abscesses. The kidneys also contained abscesses.

Laboratory Results: Mycoplasma cultures and immunohistochemistry for PI3 virus were negative. *Streptococcus* sp. and *Staphylococcus aureus* were isolated from the lung.

Contributor's Morphologic Diagnoses: 1. Severe, diffuse, chronic lymphoid interstitial pneumonia
2. Multifocal, severe, chronic suppurative bronchopneumonia with abscessation

Contributor's Comment: Ovine progressive pneumonia (OPP) and maedi-visna (from the Icelandic words for 'shortness of breath' and 'wasting')[1] are virtually identical diseases caused by closely related ovine lentiviruses. OPPV is transmitted primarily by close contact and droplet transmission and infects cells of monocyte-macrophage lineage, including dendritic cells.[2] Recent studies suggest transmission via milk and transplacental transmission are of minimal significance. Certain breeds are more susceptible to infection than others.[3] Recently, researchers in Iceland

have identified a gene (vif) in maedi-visna virus responsible for infectivity in vivo and in vitro.[4]

The gross pulmonary lesions in this case were typical of OPP complicated by secondary bronchopneumonia.[1] Heavy, grey, non-collapsing lungs often have cranioventral consolidation. Pulmonary and systemic abscessation in this ewe suggested a secondary infection with *Corynebacterium pseudotuberculosis*, although this organism was not isolated from the lungs.

The histologic lesions were diagnostic. The most characteristic feature was extensive lymphofollicular proliferations around airways and pulmonary vasculature, both arteries and veins; some had germinal center formation. Also present was the characteristic smooth muscle hyperplasia of terminal bronchioles and alveolar ducts and lymphohistiocytic interstitial pneumonia. Severe pulmonary fibrosis in the submitted sections was most likely secondary to chronic, suppurative bronchopneumonia, but 'microatelectasis' due to collapse of alveolar spaces has also been described in chronic cases.[1] Hyperplasia of type II pneumocytes and bronchiolar epithelium was not prominent, in contrast to cases of pulmonary adenomatosis, which has been associated with a type B/D retrovirus.[3] Other lesions attributed to OPPV infection seen in this ewe but not represented in these tissues were diffuse lymph node cortical hyperplasia and moderate lymphoplasmacytic synovitis in multiple joints. Also present were membranous glomerulopathy and splenic amyloidosis, which were attributed to chronic antigenic stimulation.

AFIP Diagnoses: 1. Lung: Pneumonia, interstitial, lymphohistiocytic, chronic, diffuse, severe, with perivascular and peribronchiolar lymphoid hyperplasia, and smooth muscle hyperplasia, Polypay, ovine.
2. Lung: Bronchopneumonia, suppurative, multifocal, moderate, with focally extensive abscess.

Conference Comment: Some sections contain a large abscess composed of a central area of necrosis with many small aggregates of gram-positive cocci around the periphery. This area is bounded by moderate numbers of lymphocytes, plasma cells, and fewer epithelioid macrophages. There are reactive fibroblasts and fewer inflammatory cells at the periphery.

The contributor provides a thorough overview of ovine progressive pneumonia virus disease, which is caused by a lentivirus in the *Retroviridae* family. In addition to the closely related ovine lentivirus known as maedi-visna virus, additional lentiviruses of veterinary significance include simian immunodeficiency virus (SIV),

feline immunodeficiency virus (FIV), bovine immunodeficiency virus (BIV), and equine infectious anemia virus.

The gross pulmonary lesions of uncomplicated OPP include expanded, heavy, rubbery to firm lungs that fail to collapse, and have rib impressions on the pleura. The lungs are diffusely mottled gray to grayish-tan. The bronchial and mediastinal lymph nodes are enlarged with soft grayish-white, homogeneous thickening of the cortical regions. Microscopically, the most characteristic feature of OPP is lymphocytic interstitial pneumonia with perivascular and peribronchial lymphofollicular proliferations that often have germinal centers. Other features include smooth muscle hyperplasia in the walls of terminal bronchioles and alveolar ducts, interstitial fibrosis, and microatelectasis. Hyperplasia of bronchiolar epithelium and type II pneumocytes is not a prominent feature of OPP. Other lesions associated with OPP include lymphofollicular mastitis, chronic proliferative arthritis, nonsuppurative meningoencephalitis, and vasculitis.[1]

Ovine Progressive Pneumonia (OPP) shares many clinical and pathological features with caprine arthritis-encephalitis (CAE), which is caused by a closed related caprine lentivirus. CAE virus primarily causes nonsuppurative leukoencephalomyelitis in young goats, and chronic proliferative arthritis and synovitis in adults. Less commonly, mastitis and lymphocytic interstitial pneumonia occur in adult goats CAE virus often induces two prominent pneumonic features lacking in OPP. One is extensive alveolar filling with dense, acidophilic, proteinaceous to lipoproteinaceous material, and the other is type II pneumocyte hyperplasia.[1]

Similar to goats with CAE viral pneumonia, sheep with OPP seldom have a pure viral infection, and as in this case, often develop a secondary bacterial pneumonia. It is important in such cases to separate the two processes and understand which agent is likely causing which lesion(s). When OPP is complicated by bronchopneumonia, the gross appearance should include the typical cranioventral consolidation with pus-filled airways. Additionally, there can also be coexistent lungworm lesions.[1]

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CASE II – 1140-99 (AFIP 2689105)

Signalment: 2-year-old, Quarter Horse, gelding.

History: Six, 1-2-year-old horses, out of a total of 40, died after 2-3 days of blindness and going down. One horse and another horse head were submitted. These 40 horses ate 5 tons of corn and oat screening from January 26-February 7 (our submission was on February 16). Other horses on the premises were hand-fed 5 lbs of these screenings/day/horse and were not affected.

Gross Pathology: One brain had focal, mild, yellow softening in the posterior cerebral white matter on one side. The other brain had severe yellow softening throughout the cerebral white matter, but limited to one side. There were no gross lesions in the other organs, except that the stomach was filled with roughage and finely cracked corn and oats.

Laboratory Results: Screening analysis:
Fumonisin B_1: 8.3 ppm (1.0 considered "positive" and above 5.0 ppm associated with leukoencephalomalacia) (Texas Veterinary Diagnostic Laboratory)
Aflatoxin: 5 ppb ("negative"/normal)
Rabies F.A.: negative

Contributor's Morphologic Diagnosis: Multifocally severe cerebral leukoencephalomalacia with vasculitis, hemorrhage, and gitter cells (Fumonisin toxicity).

Contributor's Comment: Equine leukoencephalomalacia (ELEM) is a neurotoxic disease caused by fumonisin production in *Fusarium moniliforme*-infected corn, usually screenings. The disease is characterized by sudden onset of circling, anorexia, head pressing, paresis, ataxia, blindness and depression that may progress to hyperexcitability. The horses usually become comatose within 1 to 10 days but can recover with some neurologic defects.

Fumonisin toxicity is cumulative so the exact "safe" level is unclear. In one study, the Fumonisin B_1 (FB_1) level never exceeded 8 ppm in horse feed not associated with ELEM, whereas the FB_1 ranged from less than 1 to 126 ppm in feed associated with ELEM with most of these being above 10 ppm. Another study indicated that 5 ppm was considered toxic. The affected horses averaged about 19 lbs/day/horse. The areas of malacia and cavitation in the cerebral white matter may be unilateral or bilateral. Microscopically, the malacic areas have gitter cells with degenerating axons and myelin and adjacent multifocal hemorrhage and slight perivascular cuffing of lymphocytes and eosinophils. Lesions occur but are less common in the spinal cord, brainstem, and cerebellum.

Fumonisins are structurally similar to sphingosine and act by blocking enzymes in sphingolipid synthesis and thus disrupt endothelial cell membranes. This might explain the vasculitis and rare fibrin thrombi in dilated vessels in this case. There is species variability in toxicity to fumonisin: horses develop neurotoxicity and sometimes hepatotoxicity, swine develop pulmonary edema, several laboratory animals develop hepatotoxicity and fumonisin is carcinogenic in rats and possibly in humans (esophageal cancer).

AFIP Diagnosis: Brain, cerebrum: Necrosis, white matter (leukoencephalomalacia), multifocal to coalescing, with vasculitis, edema, and hemorrhage, Quarter Horse, equine.

Conference Comment: There is slide variability and not all sections contain good examples of vasculitis and fibrin thrombi. Often it can be difficult to determine if there is a true vasculitis, or if the vascular changes are induced by the changes in the surrounding tissue (inflammation and necrosis). However, if fibrin thrombi are present, the vascular changes are likely not simply a result of the surrounding necrosis.

Equine leukoencephalomalacia (ELEM), also known as moldy corn poisoning, is a mycotoxicosis caused by fumonisin B_1, and is usually associated with consumption of moldy corn or grain contaminated with *Fusarium moniliforme*.[5]

In the horse, fumonisin causes two syndromes: neurotoxic and hepatotoxic. The neurotoxic syndrome is most common, with clinical signs including depression, head pressing, or seizures. Gross lesions include degeneration and liquefactive necrosis of the subcortical white matter, especially in the frontal and parietal lobes. Lesions are often bilateral, but are not always symmetrical. The characteristic gross lesion is yellow gelatinous malacia and liquefaction of the affected white matter, with hemorrhage. Microscopically, areas of liquefaction are surrounded by

diffuse or perivascular edema, hemorrhage, and small leukocytic cuffs. Blood vessels may be degenerate or necrotic with occasional thrombi. Less characteristically, there may be edema and perivascular cuffing in the leptomeninges and neuronal necrosis in the deeper layers of the gray matter. The hepatotoxic syndrome is characterized by a swollen, yellow-brown liver with multifocal pale areas. Microscopically, there is centrilobular necrosis and fibrosis that are similar to those seen with aflatoxicosis.[5]

The primary toxin isolated from *F. moniliforme* is fumonisin B_1, although other fumonisins have been extracted. The exact mechanism of injury has not been fully defined; however, vascular damage has been inferred as the primary injury. Fumonisins inhibit the enzyme ceramide synthase, interfering with the synthesis of sphingolipids. Fumonisins disrupt cellular membranes, are associated with lipid peroxidation of cells and cellular membranes, inhibit synthesis of macromolecules and DNA, and may enhance production of tumor necrosis factor-alpha by macrophages.[5]

Other animals including pigs and avian species (chickens, ducks) are susceptible, but clinical disease and lesions generally include pulmonary, hepatic, or renal injury. In pigs, the disease is called porcine pulmonary edema and is characterized by severe pulmonary edema and hydrothorax.[5] Recently, meningoencephalitis secondary to *Fusarium solani* has been reported in a German Shepherd Dog.[6]

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CASE III – A03-286 (AFIP 2948654)

Signalment: Adult (age unknown) intact male cynomolgus macaque (*Macaca fascicularis*).

History: In April 2003, the animal underwent experimental kidney transplantation and was treated with thymoglobulin and rapamycin to inhibit transplant rejection. Three weeks later the animal had labored breathing and appeared to be in pain. While sedated for a physical exam, the animal died spontaneously.

Gross Pathology: At post-mortem, the animal was thin with bilateral enlargement of the axillary and inguinal lymph nodes. There was bicavitary effusion (hydrothorax and hydroperitoneum) and the lungs had multifocal red, wet, heavy areas with intervening pink, crepitant areas. On the surface of the right kidney there was a focal, radiating white streak.

Laboratory Results: Serial Creatine values: 5/2/03 Creatine 1.9 mg/dl
5/4/03 Creatine 3.2 mg/dl
5/5/03 Creatine 3.0 mg/dl

Contributor's Morphologic Diagnoses: 1. Body as a Whole: Disseminated cytomegalovirus (CMV) infection.
2. Spleen: Severe acute multifocal necrosuppurative splenitis with intralesional CMV inclusions and CMV arteritis.

Contributor's Comment: Within the section of spleen, there is widespread loss of the white pulp denoted by severe lymphoid depletion with a conspicuous angiocentric distribution (most prominent around the central arteries). Discrete foci of intact and degenerate neutrophils admixed with bland cell detritus, karyomegalic cells and occasional small "plugs" of fibrin are predominately located within the white pulp. The karyomegalic cells are misshapen and pleomorphic with variable amounts of amphophilic cytoplasm and large oval to cigar-shaped eosinophilic glassy intranuclear inclusions surrounded by a distinct halo (Cowdry type-A inclusions). Karyomegalic inclusion-bearing cells are also scattered within the red pulp. Occasionally, endothelial cells and smooth muscle cells of large and small caliber blood vessels are karyomegalic and contain Cowdry type-A intranuclear inclusion bodies. In some sections, a focus of foreign body giant cells enveloping fragments of suture material is external to the splenic capsule.

Immunohistochemistry was performed using a polyclonal CMV antibody and cytomegalic cells show positive nuclear staining for CMV.

Cytomegalovirus (CMV) is a herpesvirus of the subfamily Betaherpesvirus and is a naturally occurring infection in many species of nonhuman primates. Following initial infection, which is often asymptomatic, the virus establishes latency in its natural host.[1] In immunocompromised animals the virus may reactivate and disseminate as the result of a variety of predisposing conditions, including immunosuppressive viral infections (SIV, type D retrovirus) and immunosuppressive drug therapy (cyclophosphamide, corticosteroids, and antithymocyte globulin).[1] Organs targeted by CMV include the eye, lung, meninges, skin, heart, intestines and testicle.[2] It is known that CMV has a predilection for mesenchymal cells, although infected cells are frequently misshapen and difficult to identify using conventional methods. The typical, almost pathognomonic cytopathological lesions produced by CMV infection are cytomegaly and large, Cowdry type-A intranuclear inclusion bodies, although both intranuclear and intracytoplasmic inclusion bodies can be seen with CMV infection.[2] In this particular animal, a satisfactory pathoanatomical explanation for the bicavitary effusion is provided by the widespread endothelial injury caused by CMV infection, resulting in increased vascular permeability and effusion into the body cavities and lungs.

AFIP Diagnosis: Spleen: Splenitis, necrotizing, acute, diffuse, moderate, with marked lymphoid depletion, and myriad cytomegalic cells with eosinophilic intranuclear inclusions, etiology consistent with cytomegalovirus, cynomolgus macaque (*Macaca fascicularis*), primate.

Conference Comment: Cytomegalovirus (CMV) is a common asymptomatic infection of humans and many nonhuman primates. CMV resembles other herpesviruses ultrastructurally, but differs from alphaherpesviruses in several aspects. First, CMV is slowly cytolytic and tends to cause enlargement of the nucleus and the cytoplasm (cytomegaly). Cytomegaly is a result of the accumulation of enveloped virions in large cytoplasmic vacuoles during viral replication instead of the virions being released into intercellular spaces. Secondly, CMV tends to be restricted in its host range, unlike many of the cytolytic alphaherpesviruses. Lastly, latent infections tend to persist in glandular tissue, lymphoreticular cells, and kidneys rather than in neurons.[1]

Cytomegalovirus is transmitted horizontally in a variety of body secretions, including saliva, blood, urine, milk, and semen. Infection is usually not associated with disease. However, disease occurs in immunocompromised individuals or following intrauterine infection. Cytomegalovirus persists as a latent infection and may periodically be shed in body secretions. In immunosuppressed macaques, reactivation of the virus may be associated with disseminated lesions in the brain, lymph nodes, liver, spleen, kidney, small intestine, nervous system, and arteries.

Disseminated CMV disease may be initiated by a variety of immunosuppressive events, including viral infection (simian immunodeficiency virus or type D retrovirus) and drug therapy (cyclophosphamide, cortisone, antithymocyte globulin).[1]

Immunosuppressive therapy is commonly given to animals and humans undergoing renal transplantation. Rapamycin is an immunosuppressive macrolide antibiotic that inhibits T and B cell proliferation, while thymoglobulin is a rabbit polyclonal antithymocyte antibody that is a potent T-cell depleting agent. Thymoglobulin is effective in depleting T-cells in both the blood and secondary lymphoid organs through the apoptotic pathway.[3]

It is important to understand the primary and secondary effects that immunosuppressive agents will have on animals when evaluating histopathological changes in specific organs. In this case, the splenic necrosis and lymphoid depletion are primary effects of the immunosuppressive agents given to this nonhuman primate. The disseminated CMV disease is likely a result of the immunosuppressive agents reducing the numbers of B and T cells, effectively diminishing this animal's immune response, and therefore allowing reactivation of the latent virus.

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CASE IV – N2004-96 (AFIP 2942031)

Signalment: 9-year-old, female, Goeldi's marmoset (*Callimico goeldii*).

History: This Goeldi's marmoset presented with an open fracture of the proximal right humerus. The tissue distal to the fracture was largely devitalized. On

physical examination the animal was icteric and had clinical signs of shock. Elective euthanasia was performed.

Gross Pathology: The carcass was in moderately thin body condition. Most tissues were discolored yellow (icterus). Both kidneys had tan mottling of the capsular surfaces as well as mottling of the medulla and cortex on cut section. The spleen was mildly enlarged, and the liver was diffusely pale tan. The open fracture of the humerus and the associated extensive devitalization of the distal right arm were confirmed.

Laboratory Results: Fluorescent antibody assay demonstrated *Leptospira* spp. antigen in kidney and liver. Microagglutination testing (MAT) (tested for 18 Leptospira serotypes) of serum obtained at the time of euthanasia revealed an antibody titer of 1:12800 for serotype Ballum.

Contributor's Morphologic Diagnoses: 1. Kidney: Nephritis, tubulointerstitial, chronic-active, diffuse, severe, with multifocal tubular regeneration, and bilirubinuria.
2. Kidney: Mineralization, tubular epithelial, multifocal, mild.

Contributor's Comment: The clinical and gross findings together with the pathological changes in the kidneys, presence of rare intratubular argyrophilic spirochetal bacteria (Steiner's silver stain), positive Leptospira fluorescent antibody (FA) result, and high antibody titer are consistent with leptospirosis in an "incidental host." In this case, infection with serotype Ballum was demonstrated.

Histologically there was, in addition to the extensive infiltration of the renal interstitium by lymphocytes, plasma cells, histiocytes, and scattered neutrophils, multifocal tubular epithelial degeneration, necrosis, regeneration, and occasionally intratubular proteinosis. Several tubules contained ongoing tubulonephritis in which infiltrating neutrophils predominated. Intratubular and intraepithelial cytoplasmic yellow-brown, granular pigment was present in several of the tubules (bilirubin pigment; Hall's bilirubin stain positive). Minimal, multifocal interstitial fibroplasia and deposition of collagen were demonstrated by trichrome staining. Few mitotic figures in cells of possible lymphocytic origin were seen in areas of inflammation. Mineralization of epithelial cells and associated epithelial degeneration/necrosis were seen in some tubules.

Leptospirosis is a zoonotic disease of global importance that affects a broad range of mammalian species. It is caused by the various serotypes of *Leptospira interrogans* (old classification scheme; see below). The bacteria are ubiquitous. Although not able to replicate in the environment, they can persist in the environment under optimal, warm and wet conditions for months.[1,2] *Leptospira* spp.

bacteria are tightly coiled spirochetes, 0.1 μm in diameter and 6-20 μm long, with hooked ends.[1] They require special methods for visualization including silver stains, fluorescent antibody methods, immunohistochemical stains, darkfield microscopy, or electron microscopy. Leptospires have also been isolated from arthropods, amphibians, reptiles, and birds, including serovars pathogenic for mammals from amphibians.[1]

"Maintenance hosts" are distinguished from "incidental hosts."[3] In the maintenance host there is a stable host-parasite relationship characterized by high susceptibility of the host species, occurrence of frequent and direct infection between individuals (usually at an early age), little or no signs of clinical disease, relatively low antibody response, and persistence of the bacteria in the renal tubules with chronic excretion via the urine. Animal species can be maintenance hosts of some serovars but incidental hosts for other serovars. Throughout the world, there are distinct variations in the maintenance hosts and the serovars they carry.[3] In domestic animals cattle may be carriers for serovar Hardjo and Pomona; pigs may carry Pomona, Tarassovi, or Bratislava; and dogs may carry serovar Canicola.[3] Examples of maintenance hosts in wild animals include raccoons and skunks in North America harboring serovar Grippotyphosa, and various species of wildlife worldwide including ungulates, harboring serovar Pomona. The most important maintenance hosts are rodents. Rats are generally thought to be carriers of serogroup Icterohaemorrhagiae serovars and mice are generally carriers for the serogroup Ballum serovars.[3] Infection with the *L. interrogans* serotype Ballum was indicated in this case by the high antibody titer.

The "incidental host" is characterized by relatively low prevalence of infection in the host population but high pathogenicity for the host, development of acute and possibly fatal disease, a short renal phase of infection with comparably short period of leptospira shedding, and a marked antibody response.[4] Possible routes of infection include contact with urine, bite wounds, and ingestion of infected tissues, as well as venereal or placental transfer.

In zoological collections, the majority of leptospirosis cases represent incidental host infections.[4] Heated buildings, that house temperature-sensitive species (i.e., tropical primates) provide an excellent, hard-to-control, year-round environment for both the Leptospira bacteria and the maintenance host; in this setting usually rodents. In this case, environmental testing to determine possible sources of infection, including water supply and the food preparation area were negative.

The mortality rate and the extent of damage to the internal organs vary with virulence of the serovar and host susceptibility; more animals encounter infections than will develop recognizable disease.[2,4] More than one serovar may occur in a given animal.[2] The bacteria penetrate either through mucous membranes or

percutaneously through injured skin. After a variable incubation period (4-20 days)[4] there is a biphasic progression of the disease; a septicemic phase lasting for about a week is followed by the immune phase. In the septicemic phase, there is leptospiremia with rapid multiplication and spread via the bloodstream. During this period, the bacteria can enter and replicate in any tissue, e.g., liver, kidney, spleen, lung, eye, central nervous tissue and genital tract.[2] Hemorrhage, intravascular hemolysis, nephritis, pneumonia, and meningitis are thought to be due to the action of bacterial toxins and the inflammatory response to the bacteria that lead to damage of the vascular endothelial lining. In some Leptospira strains hemolysins were isolated [2], some of which have been shown to mediate hemolytic and cytotoxic activities by pore formation on the mammalian cell membranes. The leptospiral lipopolysaccharide has lower endotoxic activity compared to other gram-negative bacteria.[2] Icterus, as seen in this case, is often due to a combination of intravascular hemolysis and liver injury. The spirochetes are cleared from the blood and most tissues with increasing antibody titers, but can persist and multiply for some time in renal tubules, uvea, and brain. Within the kidney, leptospires invade the interstitium with the help of their two periplasmic flagella [2] and subsequently penetrate the tubular epithelium, which results in an acute inflammatory response dominated by neutrophils. In this case, tubulointerstitial nephritis is still ongoing in some of the renal tubules, demonstrating the chronic-active nature of the inflammatory process. The acute phase of leptospirosis is transient and replaced by the immune phase, in which antibody production and excretion of leptospires via the urine are key features. Lymphocytes, plasma cells, and macrophages infiltrating the interstitium are the predominant inflammatory cells during this phase. The organisms can persist within the renal tubules of the incidental host, protected from antibody and other host defenses, for a few days to several weeks. Renal insufficiency and failure are associated with the tubular damage, and possibly with decreased glomerular filtration and hypoxia due to the overall organ swelling that may impair renal blood perfusion.[2] If infection takes place during pregnancy, fetal infection may occur, resulting in abortion (usually last trimester), stillbirth, birth of weak neonates, or birth of healthy but infected neonates.[4] Detectable gross lesions are often absent in infected neonates; thus, failure to further test for leptospira may result in under-reporting of the disease.[5]

In the old phenotypic classification system, the *Leptospira* were serologically divided into two species. *Leptospira interrogans* included all the pathogenic serovars, and *Leptospira biflexa* included all the environmental, nonpathogenic serovars.[3] The more recent classification, based on genotypic properties (12 named and 5 unnamed genomospecies), is somewhat problematic in the clinical situation, since it is incompatible with the former system of serogroups (e.g., species in the new system do not correspond to the previous two species and their serovars, and include pathogenic and nonpathogenic within some species).[3] Therefore, the

phenotypic classification is likely to remain in place until simpler molecular identification methods are developed.[3]

Fluorescent antibody (FA) testing is frequently used to confirm the presence of leptospiral antigen in fresh urine or fresh or frozen tissue collected at the time of necropsy.[5] If serum is available, microagglutination testing (MAT) may help to determine the infecting serovar. The highest titer is considered the infecting serovar, with lower positive titers considered to be cross-reactivity.[2] Leptospires can be isolated from blood and CSF during the first phase of illness, and from urine after the first week (data from humans);[3] overall, culture is problematic due to the fragile and fastidious nature of the organisms.[2,5] The use of PCR assays is also still limited due to the inability of most assays to identify the infecting serovar.[3] Killed leptospiral vaccines are frequently used in domestic species and occasionally in exotic species such as the black rhinoceros, in which leptospira infections have been associated with hemolytic crises and high mortalities.[4]

AFIP Diagnosis: Kidney: Nephritis, interstitial, lymphocytic, multifocal, moderate, with neutrophilic tubulitis, Goeldi's marmoset (*Callimico goeldii*), primate.

Conference Comment: The contributor provides a very thorough overview of leptospirosis in animals, including the epidemiology, transmission, histopathological changes, and diagnostic techniques.

Prior to being informed of the contributor's laboratory results, some conference attendees considered lymphoma as a primary differential diagnosis based on the relatively monomorphic population of interstitial lymphocytes that are associated with moderate numbers of mitotic figures, and in some slides the apparent blurring of vessel walls by high numbers of lymphocytes.

Although the angiotropic lymphomas were considered, there are several histopathologic changes for which one must account. First, although the infiltrating inflammatory cells are predominantly lymphocytes, there are low numbers of plasma cells and histiocytes within the interstitium. Therefore, there is not truly a monomorphic population. Secondly, lymphoma classically forms sheets of cells that obliterate normal tissue architecture and often induce a mass effect; neither is evident in this section. And, although the angiotropic lymphomas could be considered, the lymphocytes are not predominantly associated with vessels, nor are they occluding vessels resulting in ischemic infarction. Lastly, one must account for the renal tubular inflammation, degeneration, and necrosis.

When considering all of the changes apparent with H&E alone: tubular degeneration and necrosis, neutrophilic tubulitis, and lymphoplasmacytic and histiocytic interstitial nephritis, the most logical differential is leptospiral nephritis.

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