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CASE I – 04-113 (AFIP 2942979)

Signalment: 9-year-old, neutered male, Rottweiler/Shepherd dog mix.

History: The dog had a one-month history of "not doing right". On presentation, lethargy and vomiting had developed. The clinician noted anemia (PCV – 27%) and increased liver enzymes. On physical exam a palpable abdominal mass was noted and by imaging was determined to be an enlarged spleen. Peripheral lymph nodes were not enlarged. On exploratory laparotomy, the spleen was confirmed to be extremely large with irregular borders and a mottled surface. A splenectomy was performed and the spleen submitted for histopathology exam. The left lateral and medial lobes of the liver appeared mildly enlarged with rounded edges. No biopsy was taken of the liver. No other gross abnormalities were found within abdomen.

Gross Pathology: The spleen was markedly, diffusely enlarged (approximately 2-3 times normal size) and firm.

Laboratory Results: CBC day of surgery:

CBC	Patient Value	Reference Range	Units	Percentage
WBC	66	5.5 – 16.9	X $10^3/\mu l$	
Neutrophils	7.06	2.0 – 12.0	X $10^3/\mu l$	10.5
Monocytes	19.28	0.1 – 1.4	X $10^3/\mu l$	28.8
Lymphocytes	39.48	0.7 – 4.9	X $10^3/\mu l$	58.9
Basophils	0.36	0.0 – 0.1	X $10^3/\mu l$	0.5
Eosinophils	0.81	0.1 – 1.49	X $10^3/\mu l$	1.2
RBC	2.72	5.5 – 8.5	X $10^6/\mu l$	
Hb	7.2	12.0 – 18.0	g/dl	
Hct	21.6	37.0 – 55.0	%	
Reticulocytes	147.3		X $10^3/\mu l$	5.4
MCV	79.5	60.0 – 77.0	Fl	
MCHC	34.1	31.0 – 37.0	g/dl	
MCH	27.07	19.5 – 24.5	Pg	
RDW	16.3	14.7 – 17.9	%	
PLT	154		X $10^3/\mu l$	
MPV	20.38		Fl	
PCT	0.3		%	
PDW	20.9		%	

Contributor's Morphologic Diagnosis: Spleen: T-cell LGL Lymphocyte lymphoma/leukemia, Rottweiler/Shepherd mix, canine.

Contributor's Comment: The splenic red pulp is diffusely expanded by numerous medium to large neoplastic round cells with scant amounts of eosinophilic cytoplasm and round to indented nuclei. Scattered germinal centers in the white pulp are still evident but are mildly atrophic. Minimal extramedullary hematopoiesis is present. Immunohistochemical staining demonstrates neoplastic cells to be strongly positive for CD3 and negative for CD79a and lysozyme.

Venous blood smears reveal a leukemic blood profile with a moderate regenerative anemia. Neoplastic lymphocytes are characterized by medium to occasionally large cells with moderate amounts of basophilic cytoplasm, round to irregularly indented nuclei with moderately clumped chromatin and variably visible nucleoli. Many cells have low numbers of variably sized, azurophilic granules that are often perinuclear. Occasional larger blast cells are noted. A rare mitotic figure is present. A manual WBC differential count on the blood smear results in 97% of cells being neoplastic lymphocytes, with 2% segmented neutrophils and 1% eosinophils. When compared to the automated differential count, it is apparent many of the neoplastic lymphocytes were erroneously classified as monocytes by the hematology analyzer.

Large granular lymphocyte (LGL) leukemias/lymphomas are either of cytotoxic T-cell (CD8+CD3+) or natural killer (NK) (CD3-) cell origin.[1] LGLs have characteristic azurophilic granules in their cytoplasm often in a perinuclear location; ultrastructurally, these granules appear as membrane-bound structures with an electron-dense core. In normal dogs up to 10% of peripheral lymphocytes in the blood can be LGLs.[2] Unlike acute lymphocytic leukemias, which originate in the bone marrow, canine LGL lymphoma/leukemias are thought to originate from $\alpha_d\beta_2^+$ lymphocytes in the red pulp of the spleen.[3]

In addition to surface antigens, cytotoxic granule proteins such as TIA-1, granzyme, and perforin are used immunologically and in molecular studies to identify LGL leukemic cells in people.[4] Perforin-like immunoreactivity has been demonstrated in feline LGL lymphomas[5] as well as rats, mice, and guinea pigs.[2]

LGL lymphoma/leukemia in some dogs can be an aggressive disease involving spleen, liver, and bone marrow. Leukemic blood profiles with lymphocyte counts of up to 138 x 10^6/ml have been reported, with the majority (80-97%) being LGLs. In other dogs, the disease progress is slower, behaving like chronic lymphocytic leukemias.[2] LGL lymphoma in cats is usually associated with the gastrointestinal tract and tends to be an aggressive disease that spreads rapidly to the spleen, liver, and lymph nodes.[5, 6] Leukemia is variably seen in cats. LGL lymphoproliferative disorders have also been reported in horses[7], ferrets, birds,[2] and is common in Fischer 344 rats. Non-neoplastic proliferations of LGL in dogs have been reported with chronic ehrlichiosis.[3]

In humans, LGL leukemia is thought to arise from apoptosis dysregulation due to abnormalities in the Fas/Fas ligand pathway.[8] LGL leukemia cells are resistant to Fas-mediated apoptosis and express high levels of both Fas and Fas ligand.[8] Multiple genes involved in cytotoxic functions are upregulated in LGL leukemia including granzymes, cathepsin, calpain, perforin, and capase-8, similar to that seen in activated cytotoxic T-cells.[8] LGL lymphoma/leukemia is often associated with autoimmune disorders, particularly rheumatoid arthritis, and other lymphoproliferative disorders. Clonal proliferations of LGLs in humans have been associated with Epstein-Barr virus, human immunodeficiency virus, and human T cell lymphotropic virus.[2] Type C oncovirus has been reported associated with a cell line derived from peripheral lymphocytes from a dog with LGL leukemia.[9]

AFIP Diagnoses: 1. Spleen, sinusoids: Large granular lymphocyte leukemia, mixed breed, canine.
2. Spleen, white pulp: Lymphoid atrophy, diffuse, moderate.

Conference Comment: Some sections contain discrete nodules of white pulp that are unaffected by the neoplastic cells suggesting this neoplasm originates from the red pulp rather than the white pulp. It can be difficult (if not impossible) to differentiate lymphoma from leukemia when there is advanced disease with infiltration of neoplastic cells into widespread tissues. In these instances, special methods such as immunophenotyping of the cells are necessary to help differentiate the conditions.

Lymphoma is a neoplasm of lymphocytes arising as a solid tissue mass in organs other than bone marrow.[10] Lymphoma can be classified into subtypes according to anatomic distribution, histiologic pattern, cellular morphology, cytochemistry, and expression of cluster designation (CD) markers. Subtypes of lymphoma according to distribution include: multicentric, alimentary, mediastinal, cutaneous, and miscellaneous. Multicentric lymphoma is the most common type of lymphoid neoplasia in dogs, cattle, and horses, while alimentary lymphoma is the most common form in cats. The histologic pattern in most cases is diffuse and characterized by sheets of neoplastic lymphocytes that efface and replace normal tissue architecture.[11]

Leukemia is defined as hematopoietic neoplasia with neoplastic cells in the blood and/or bone marrow. Lymphocytic leukemia arises from the bone marrow and may be either acute or chronic. Acute lymphocytic leukemia (ALL) is most common in younger animals, arises from undifferentiated lymphocytes in the bone marrow that are often of B-cell origin, and usually presents as large blastic cells. Chronic lymphocytic leukemia (CLL), is more common in older animals, arises from relatively differentiated lymphocytes that "home" to secondary lymphoid organs such as the spleen. These cells resemble small lymphocytes and are often of T-cell origin, specifically the CD8+ subset.[11]

There are several accepted classification schemes for hematopoietic tumors, including the REAL (Revised European and American Lymphoma) system and WHO (World Health Organization) system. The REAL system categorizes tumors based on their histogenetic derivation and biological behavior, while the WHO system also includes acute and chronic myeloproliferative diseases as well as myelodysplastic syndromes. Although the classification schemes may appear complex, the differentiation of specific hematopoietic tumors is important for veterinary oncologists to provide optimal tumor management.[1]

The contributor provides a thorough overview of LGL lymphoma/leukemia in domestic animals and humans. Although the azurophilic granules are difficult to appreciate on the H&E slides, granules can be seen in lymphocytes particularly around the fibromuscular trabeculae. When LGL lymphoma/leukemia is suspected,

it is often easier to diagnose using cytologic touch imprints of affected organs or peripheral blood smears.

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CASE II – N04-95 (AFIP 2937501)

Signalment: 3.5-year-old, female, French Alpine goat, *Capra hircus*, caprine.

History: This doe was presented with a 2-month period of dyspnea and progressive emaciation after parturition. The animal was euthanized and necropsied.

Gross Pathology: The doe was in poor body condition and exhibited minimal fat stores. There were multifocal fibrous adhesions between the parietal and visceral layers of the pleura (Fig. 1). Both lungs exhibited multifocal, green yellow nodules (abscesses) ranging from 0.5-2 cm in diameter. These nodules were primarily located in the cranial lobes. The abdominal cavity contained approximately 1 litre of clear, light yellow watery fluid (ascites). The liver showed multifocal confluent nodules ranging from 0.5-5 cm in diameter. Abundant pale green viscous material (pus) oozed freely from the nodules on cut surface (Fig.2). The rumen had a focal, approximately 1.5 cm in diameter, area of hemorrhage located in the serosal surface near the cardia. At this site, the mucosa exhibited blunting and erosion of the ruminal papillae. The mesenteric lymph nodes were enlarged twice their normal size. Both kidneys displayed numerous, miliary white foci scattered throughout the cortex. The rest of the internal viscera were unremarkable.

Laboratory Results: An hour previous to euthanasia, a complete blood count revealed a leukocytosis, neutrophilia, and mild monocytosis associated with chronic active inflammation. A biochemical profile revealed azotemia, hyperproteinemia, hyperglobulinemia, hypoalbuminemia, hypocalcemia, hyperkalemia, hyponatremia, and hypochloremia and elevated GGT and CK.

CBC/Chem	Patient Value	Reference Range	Units
WBC	30.2	4.0 – 13.0	X 10^9/L
Neutrophils	22.7	1.2 – 2.7	X 10^9/L
Monocytes	0.6	0.0 – 0.55	X 10^9/L
BUN	19.9	3.5 – 6.66	mmol/L
Creatinine	643	97 – 159	mmol/L
Protein	87	59 – 76	g/L
Globulin	78	20 – 60	g/L
Albumin	9	27 – 39	g/L
Calcium	1.08	2.22 – 2.91	mmol/L
Potassium	7.82	3.5 – 6.7	mmol/L
Sodium	127	142 – 155	mmol/L
Chloride	94	99 – 110	mmol/L
GGT	60	< 56	U/L
CK	3351	< 8.9	U/L

Bacteriology: A heavy growth of *Arcanobacterium pyogenes* was isolated in pure culture from the lung and liver lesions.

Parasitology (fecal flotation): *Eimeria* spp

Contributor's Morphologic Diagnosis: 1. Liver: Multifocal confluent abscesses.
2. Liver: Hepatitis, necrosuppurative, plasmacytic and histiocytic, multifocal, severe, chronic, with myriads of intralesional bacteria, and abundant, diffuse, intercellular, intrahistiocytic, radiating to homogeneous, eosinophilic material consistent with Splendore-Hoeppli material.
3. Kidney: Glomerulonephritis, proliferative, neutrophilic, fibrinous, global, diffuse, severe, with glomerular hemorrhage, tubular dilation, proteinosis and degeneration, and scant, multifocal, extracellular, radiating to homogeneous, eosinophilic material consistent with Splendore-Hoeppli material.
4. Kidney: Nephritis, tubulointerstitial, neutrophilic and lymphoplasmacytic, multifocal, severe, subacute.
5. Lung: Multifocal abscesses and fibrous pleural adhesions (slides not submitted).
6. Rumen: Rumenitis, necrosuppurative and hemorrhagic, focal, acute, moderate (slides not submitted).

Contributor's Comment: The section of liver shows large, multifocal areas of liquefactive necrosis containing myriad bacteria intermixed with numerous degenerate neutrophils and cellular debris. These areas are surrounded by large numbers of plasma cells, with lesser numbers of histiocytes, neutrophils and rare lymphocytes. Multifocal aggregates of neutrophils, plasma cells and lymphocytes are scattered throughout the parenchyma. The presence of abundant, homogeneous, eosinophilic, hyaline material often with radiating projections (spiculated), diffusely distributed among the parenchymal cells and sometimes within Kupffer cells is remarkable. This material ranges approximately from 20 to 30 microns in diameter, is usually surrounded by moderate numbers of neutrophils and is considered as consistent with Splendore-Hoeppli substance. Same material was ruled out as amyloid through Congo red staining (not submitted).

The section of kidney exhibits increased cellularity of the glomerular tufts caused by proliferation of epithelial and mesangial cells, with hypertrophy of the epithelium and focal adhesions between the glomerular tuft and Bowman's capsule (synechiae). Most of the glomeruli have small clusters of neutrophils and small deposits of fibrin involving both the glomerular tufts and the urinary space. In some urinary spaces the inflammatory cells are mixed with abundant red blood cells. Most of the tubules exhibit marked dilation and contain abundant, intraluminal, pale eosinophilic, proteinaceous material. The tubular epithelium has multifocal vacuolar degeneration with occasional intracytoplasmic hyaline droplets. Multifocal small aggregates of neutrophils, plasma cells and lymphocytes are

scattered throughout the interstitium. There are also numerous intraluminal clusters of neutrophils mainly located in the medullary region. Occasional deposits of Splendore-Hoeppli material are present in some glomerular tufts.

The order Actinomycetales includes several families of pathogenic organisms. These organisms, referred to as higher bacteria, often generate lesions that resemble those produced by fungi. The genera *Actinomyces, Nocardia, Rhodococcus, Corynebacterium, Dermatophilus, Streptomyces,* and *Mycobacterium* are included in this order.[1] *Arcanobacterium pyogenes*, formerly known as *Corynebacterium pyogenes* and *Actinomyces pyogenes*, is widespread throughout the word as a common cause of pyogenic processes in cattle, sheep, swine, goats, and wild ungulates.[1,2] It is a facultatively anaerobic, small, pleomorphic, gram-positive bacterium, which is considered a normal inhabitant of the mucous membranes of several domestic species. *Arcanobacterium pyogenes* can induce purulent inflammation almost anywhere in the body. Usually this takes the form of localized abscesses, but the lesions may be more diffuse in internal viscera, joints, or tendon sheaths.[2] In goats, purulent pneumonias and abscesses in the upper respiratory tract,[1] as well as mandibular osteomyelitis[3] have been described as lesions associated with *A. pyogenes* infection.

A distinctive morphologic feature of certain fungal and bacterial diseases is the presence of a homogeneous, brightly eosinophilic substance, known as Splendore-Hoeppli material, surrounding individual organisms or colonies of organisms. It exists as a surrounding collar of radially arranged clubs, or as a variably-sized rim often with a serrated edge. The exact nature of this material is unknown, but it appears to be a product of the host, most likely antigen-antibody complexes. Splendore-Hoeppli material is a relatively consistent feature of coccidioidomycosis and sporotrichosis, as well as certain diseases caused by higher bacteria, such as actinomycosis.[4] Other bacterial infections associated to this phenomenon are those produced by *Actinobacillus lignieressi*[5] and *Staphylococcus aureus* (Botryomycosis).

To our knowledge, the Splendore–Hoeppli phenomenon is not a common histomorphologic feature in infections caused by *Arcanobacterium pyogenes*. However, it was a remarkable finding in the liver section of this goat. It is also interesting that the material described in this case was not surrounding bacterial colonies, as it has been described in the literature.

AFIP Diagnoses: 1. Liver: Abscesses, multifocal and coalescing with myriad bacilli and, moderate, random, portal, neutrophilic and plasmacytic hepatitis, with abundant eosinophilic spiculated material, French Alpine goat, caprine.
2. Kidney: Glomerulonephritis, necrotizing, hemorrhagic and neutrophilic, global, diffuse, severe, with moderate multifocal, neutrophilic and plasmacytic, tubulointerstitial nephritis, numerous fibrin thrombi, and multifocal eosinophilic spiculated material.

Conference Comment: Within the liver sections, two prominent features are present, the focal areas of necrosis with large colonies of bacteria and the abundant eosinophilic spiculated material.

There are several gram-positive and gram-negative bacteria that form large colonies in tissue. They are known by the pneumonic YAAACCSS[5,6,7,8]:

Yersinia sp.	Gram-negative
Actinomyces sp.	Gram-positive
Actinobacillus sp.	Gram-negative
Arcanobacterium sp.	Gram-positive
Clostridium sp.	Gram-positive
Corynebacterium sp.	Gram-positive
Staphylococcus sp.	Gram-positive
Streptococcus sp.	Gram-positive

There is abundant eosinophilic, often spiculated material surrounding areas of necrosis and within sinusoids in unaffected areas. Conference attendees discussed whether this could be fibrin thrombi, Splendore-Hoeppli material or amyloid. Fibrin thrombi often create a long cast of the vascular lumina and tend to contain more enmeshed erythrocytes. PTAH (Phosphotungstic Acid Hematoxylin) stains fibrin dark blue, but did not stain the material. The spiculated morphology is most consistent with Splendore-Hoeppli material. Although the exact nature of Splendore-Hoeppli material is not known, it is thought to be antigen-antibody complexes. These complexes are often found closely associated with the bacteria. In this case, a significant amount of the material was not associated with bacteria and often not associated with inflammation. Tissue Gram stains did not reveal bacteria within the eosinophilic material. Amyloid is a pathologic proteinaceous substance that histologically appears as an amorphous, eosinophilic, hyaline, extracellular material. Within the liver, amyloid first appears in the space of Disse, and with progressive accumulation, produces pressure atrophy of adjacent cells. It is congophilic, and exhibits green birefringence when polarized. After staining with Congo Red locally, the eosinophilic material is multifocally congophilic with green birefringence. There are several forms of amyloid, but the most common are AL (amyloid light chain) and AA (amyloid associated). AL protein is composed of partial or complete immunoglobulin light chains and is most commonly associated

with B cell/plasma cell dyscrasias. AA protein is produced in the liver from SAA (serum amyloid-associated), which is an acute phase protein, and is most commonly seen in association with chronic inflammatory disorders.[9] Without further testing, it is not possible to definitively determine the composition of the eosinophilic material in this case.

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CASE III – 04-437 (AFIP 2933948)

Signalment: 3-year-old, intact female, Duncan Hartley (strain HsdPoc:DH) guinea pig *(Cavia porcellus)*

History: This colony guinea pig was a short-haired albino with a recent history of muscle mass loss and a palpable intra-abdominal mass. There had been no recent experimental manipulations or breeding; this group of guinea pigs was kept mainly for environmental enrichment. The guinea pig was found dead and presented for necropsy on the same day.

Gross Pathology: At necropsy, there was marked muscle thinning on the cervical, thoracic and abdominal regions. The lungs were diffusely firmer than normal and the sternal lymph nodes were wet and red. The liver was diffusely red/brown, friable and granular and the spleen contained a 1 cm diameter soft, red nodule. Both kidneys had a granular appearance. Both ovaries were markedly enlarged by fluid-filled, thin walled cysts, probably accounting for the abdominal mass palpated prior to death.

Laboratory Results: None performed

Contributor's Morphologic Diagnosis: 1. Liver, lung: Cavian leukemia.
2. Liver: Mild centrilobular hepatocellular fatty change.

Contributor's Comment: Microscopically, the hepatic sinusoids were expanded by a fairly monomorphous population of round, lymphoid cells with scant to small amounts of pale eosinophilic cytoplasm and centrally placed euchromatic to hypochromatic, faintly stippled nuclei. Nuclei ranged from oval to round or indented; nucleoli were inconspicuous. Mitotic figures averaged 6 per 10 hpf (40X) and scattered tingible body macrophages created a "starry sky" effect, particularly in portal areas, where the lymphoid cells were more numerous and solidly packed. The capsular surface was a little irregular due to the subcapsular accumulation of lymphoid cells. Larger veins throughout the liver contained similar round cells. Mild, centrilobular hepatocellular fatty change was noted. In the lung, there was diffuse interstitial thickening due to the presence of lymphoid cells similar to those described above. These also occupied larger, congested veins. Incidental foci of heterotopic bone were also present.

Other infiltrated organs included lymph nodes (although peripheral lymphadenopathy was not appreciated at gross necropsy), the spleen, kidneys, intestinal tract, adrenal and thyroid glands. Most blood vessels contained similar cells. Bone marrow cellularity was 95-100% but the cells were still mixed, although there was a predominance of larger, non-segmented cells. Due to

decalcification, it was difficult to be certain if these were the same population as was present throughout the rest of the body. However, reportedly there is lesser involvement of the bone marrow.[1]

Cavian leukemia is a spontaneous form of hematopoietic neoplasia that can occur in inbred (strains 2/N and 13/N) and non-inbred lines. Disease tends to arise in young adults under three years of age. The incidence varies in different reports, ranging from under 0.01% in a 16-year period[2] to 0.2% over a three-year period.[3] Early experimental work[4] demonstrated transmission of a "leukemogenic agent" via cell-free inoculation or cell transplantation to hybrid guinea pigs. Electron microscopy revealed Type C virus particles in neoplastic lymphoid cells, suggesting a role for retrovirus infection. Experimentally, this is also a 100% transmissible (transplantable) form of neoplasia in inbred strains and some hybrids (e.g. strain 2/Hartley), usually by intraperitoneal or subcutaneous inoculation of fresh or thawed frozen whole blood or tissue. This has allowed maintenance of leukemic lines and further study of the pathogenesis, although it is not entirely clear if more recent spontaneous cases reflect exactly the same disease process.[2] The degree of transmissibility decreases quickly with age in non-inbred strains but can be facilitated by prior corticosteroid treatment.[1] While this disease is typically thought of as retrovirus-associated, guinea pig herpes-like virus may also contribute to the development of the leukemia. Herpes-like virus particles have been isolated from lymphoid cells derived from leukemic guinea pigs but only *in vitro*[5] and the cells were also derived from guinea pigs infected with retrovirus. Cavian leukemia has been postulated as a model for acute lymphoblastic leukemia in humans.[1]

AFIP Diagnosis: Lung; liver: Leukemia, lymphoblastic, Duncan Hartley guinea pig, cavian.

Conference Comment: Cavian leukemia is a rare condition that occurs spontaneously in various inbred and non-inbred strains of guinea pigs. Affected animals are usually young adults and may present with either a generalized lymphadenomegaly and/or a leukocytosis (up to 180,000 mm^3) with a significant number of circulating lymphoblastic cells. At necropsy, in addition to enlarged peripheral lymph nodes, there is marked splenomegaly and hepatomegaly. Histologically, as was noted in this case, there is diffuse infiltration of lymphoblastic cells in the liver, interstitium of the lung, as well as, spleen, bone marrow, thymus, alimentary tract lymphoid tissue, heart, eyes, and adrenal glands. As the contributor mentions, an endogenous retrovirus, is associated with, but not proven to cause Cavian leukemia. Although a retrovirus appears to play an important role in this disease, C-type virus particles have been noted in lymph node germinal centers from normal guinea pigs.[6]

Many viruses have been associated with tumor induction in both animals and humans. Most of these are DNA viruses and include the following[7]:

Family/Genus	Virus	Associated Tumor
DNA Viruses		
Poxviridae		
Leporipoxvirus	Rabbit fibroma virus	Myxoma
	Squirrel fibroma virus	Fibroma
Yatapoxvirus	Yaba monkey tumor virus	Histiocytoma in monkeys
Herpesviridae		
Alphaherpesvirinae	Marek's disease virus	Lymphoma in fowl
Gammaherpesvirinae	Ateline herpesvirus-2	Lymphoma in
	Saimirine herpesvirus-2	Lymphoma in
	Epstein-Barr virus	Lymphoma in monkeys
	Baboon herpesvirus	Lymphoma in baboons
	Cottontail rabbit herpesvirus	Lymphoma in rabbits
Ungrouped	Lucké frog herpesvirus	Renal adenocarcinoma
Adenoviridae		
Mastadenovirus	Many adenoviruses	Solid tumors in rodents
Papovaviridae		
Papillomavirus	Cottontail rabbit papillomavirus	Papillomas in rabbits
	Bovine papillomavirus-4	Papilloma, carcinoma of intestine/urinary bladder
	Bovine papillomavirus-7	Papilloma, carcinoma of the eye
	Human papillomavirus-5, 8	SCC
	Human papillomavirus-16, 18	Genital carcinoma
Polyomavirus	Murine polyomavirus	Solid tumors in rodents
Reverse Transcribing Viruses		
Hepadnaviridae (DNA virus)		
Orthohepadnavirus	Woodchuck hepatitis virus	Hepatocellular carcinoma
	Human hepatitis virus	Hepatocellular carcinoma
Avihepadnavirus	Duck hepatitis virus	Hepatocellular carcinoma

Family/Genus	Virus	Associated Tumor
Retroviridae (RNA virus)		
Alpharetrovirus	Avian leukosis virus	Lymphoma/leukemia
	Rous sarcoma virus	Sarcoma in fowl
	Avian erythroblastosis virus	Erythroblastosis in fowl
	Avian myeloblastosis virus	Myeloblastosis in fowl
Betaretrovirus	Mouse mammary carcinoma virus	Mammary carcinoma
	Mason-Pfizer monkey virus	Sarcoma and Immunodeficiency
Gammaretrovirus	Feline leukemia virus	Leukemia
	Feline sarcoma virus	Sarcoma
	Murine leukemia virus	Lymphoma/leukemia
	Murine sarcoma virus	Sarcoma
	Avian reticuloendotheliosis Virus	Reticuloendotheliosis
Deltaretrovirus	Bovine leukemia virus	Leukemia
	Jaagsiekte virus	Adenocarcinoma of the lung in sheep
	HTLV-1 and –2	Human adult T cell leukemia and hairy cell leukemia
	Simian HTLV virus	Leukemia in monkeys
RNA Viruses		
Flaviviridae		
Hepacivirus	Hepatitis C virus	Hepatocellular carcinoma in humans

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CASE IV – R 18 (AFIP 2937814)

Signalment: 13-month-old, male, Cavalier King Charles Spaniel.

History: A porto-azygos shunt was diagnosed by the veterinarian and 2 surgical interventions were undertaken within an 11 week interval.
Before the second intervention, the basal portal pressure was 9 mm Hg. After the ligature, it was 24 mm Hg and euthanasia was elected.

Jan 29th 2004, convulsive seizure:
Abdominal echography: very small liver, porto-azygos shunt, cholelithiasis, increased kidney size.

March 18th 2004, first surgery

June 2nd 2004, echography
Early signs of portal hypertension and persistent shunt.
Anorexia, polyuria polydipsia, convulsive seizure.

June 10th 2004, second surgery
Ascites
Euthanasia

Gross Pathology: Porto-azygos shunt, small-sized liver

Laboratory Results: Jan 29th 2004, convulsive seizure:
Alkaline phosphatases: 495 UI
ALAT: 540 UI
Pre-prandial bile acids: 110 mmol/L
Post-prandial bile acids: 261 mmol/L

Contributor's Morphologic Diagnosis: Liver: Arteriolar hyperplasia, portal, diffuse, moderate, with terminal hepatic vein hypoplasia, lobular atrophy, periportal and bridging fibrosis, consistent with congenital portosystemic vascular shunt.

Contributor's Comment: The liver architecture is modified, as the hepatic lobules are difficult to identify due to loss of hepatocytic plates and terminal hepatic veins. The lobules seem small as evidenced by a subjective decrease in distance between portal triads, and there is slight periportal and bridging fibrosis. Serpentine arrangements of arteriolar smooth muscle cells that are disposed in a perilobular pattern represent portal arteriolar hyperplasia. Strikingly, portal veins are still discernible. Hepatocytes are small, dissociated, and a proportion of them show some medium-sized vacuoles. Ito cells are abundant and show a large and unique vacuole. Brown pigment is found in scattered Kupffer cells and bile canaliculi (cholestasis).

Portosystemic shunts are congenital or acquired communications between the portal and systemic vasculature.[4] Congenital vascular shunts are described more often in dogs, less frequently in cats, and sporadically in other domestic animals.[1] Rarely, young dogs may have arteriovenous (arterioportal) fistulae and develop portal hypertension, ascites, and acquired shunts.[3]

Clinically, the animals may be small for their age and show neurological signs due to hepatic encephalosis resulting from inadequate clearance of enterically derived toxins in portal blood.[2] In addition to neurological signs, animals with shunts may suffer from renal, cystic, or urethral calculi due to increased urinary excretion of ammonia and uric acid. The formation of ammonium biurate crystals in urine is frequent.

Biologically, increased post-prandial bile acids are typical of this condition. Dogs with portosystemic shunts often have hypoalbuminemia in the absence of proteinuria, low blood urea nitrogen, hypoglycemia, and hypocholesterolemia due to decreased hepatic function. Erythrocytic microcytosis is a common finding in animals with portosystemic shunts although iron metabolism is normal.[1]

AFIP Diagnosis: Liver: Portal arteriolar hyperplasia and venule hypoplasia, diffuse, moderate, with lymphangiectasia, hepatocellular atrophy and fatty change, periportal and bridging fibrosis, and bile stasis, Cavalier King Charles Spaniel, canine.

Conference Comment: The blood flow to the liver is unique with the hepatic artery providing oxygenated blood, the portal vein providing blood flow from the intestinal tract and spleen, and the hepatic vein returning blood from the liver to the systemic circulation. In health, portal blood contains constituents absorbed from the intestinal tract, including bile acids, amino acids, glucose, ammonia, medium-length fatty acids, and intestinal antigens that are largely removed by the liver before they reach systemic circulation. In acquired or congenital shunts, the portal blood largely bypasses the liver, and directly enters the systemic circulation. Therefore, systemic blood concentrations of the substances normally removed by hepatic processing are increased (i.e. bile acids, ammonia). Hyperammonemia may lead to CNS signs (hepatic encephalopathy).[5]

Portosystemic shunts often result in hepatic atrophy, often with concomitant loss of functional mass, due to decreased concentrations of intestinal and pancreatic hepatotrophic factors that normally reach the liver through the portal circulation. Approximately 70% or more of the functional hepatocytes must be lost before alterations of hepatic function are detectable by serum chemistry. When the functional mass is significantly reduced, it may result in hypoproteinemia, hypoalbuminemia, hypoglycemia, hypocholesterolemia, decreased BUN, and hyperbilirubinemia.[5]

The clinical pathology findings in this case are classic for animals with portosystemic shunts. The fasting and postprandial bile acids are elevated due to the portal vein largely bypassing the liver and delivering the blood to the systemic circulation. The increased ALAT (ALT, alanine aminotransferase) indicates hepatocellular injury with enzyme leakage from the cytosol of the hepatocytes into the blood. ALP (alkaline phosphatase) is an inducible hepatic enzyme with several isoenzymes, including liver, corticosteroid, bone, intestinal, and placental. These isoenzymes can be differentiated by their electrophoretic mobility. The liver ALP isoenzyme is a sensitive indicator of intrahepatic or extrahepatic cholestasis, whereas the bone ALP isoenzyme is frequently observed in young, rapidly growing animals. In this case, both isoenzymes may be contributing to the elevated ALP enzymatic activity.[5]

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