Joint Pathology Center
Veterinary Pathology Services

WEDNESDAY SLIDE CONFERENCE
2020-2021

Conference 3

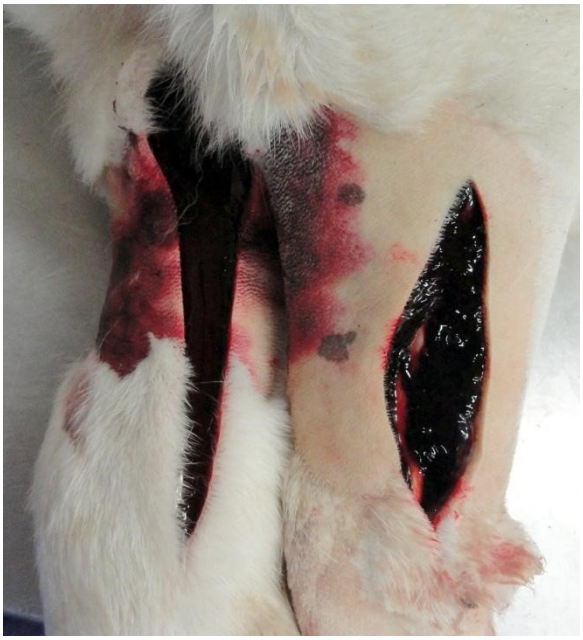
2 September, 2020

Joint Pathology Center
Silver Spring, Maryland

CASE 1: 11240-12 (4032246-00)

Signalment: 1.5-year-old female St. Bernard dog (*Canis lupus familiaris)*

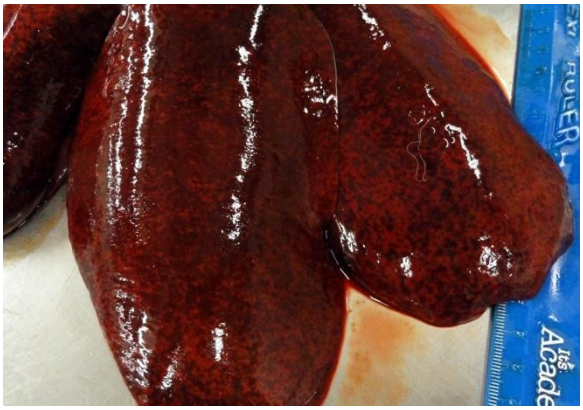
History: The owner had taken 3 dogs to a show two weeks previously and they subsequently developed clinical signs of coughing. All dogs (all related St. Bernards) were then treated with trimethoprim sulfa at an unknown dose, over a ten-day period. This dog presented with seizures and hypoglycemia. There was severe anemia, thrombocytopenia, and dry eye. Two other littermates with similar clinical signs and similar treatment were hospitalized, treated symptomatically and eventually recovered. Drug treatment was withdrawn from the other dogs one day following this dog's necropsy.



Lower legs, dog: There is extensive subcutaneous hemorrhage (Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, 1600 East Rollins Street, Columbia MO 65211)

Gross Pathology: This young adult dog had body weight 71.8 kg. The dog was moderately autolyzed. Fecal material on the perineal hair had a red hue. Beginning in the mid-jejunum, intestinal content was dry and consisted of blood. The lower legs were swollen with what was believed to be pitting edema. This was found to be a result of 1-3 cm thick subcutaneous hemorrhage.

Additional pockets of subcutaneous hemorrhage were present over the rib cage and in the left jugular groove. Dry mucus was observed in the left conjunctiva. The spleen was enlarged and weighed 0.47% body weight (the animal was euthanized. The liver was 1.8% body weight (normal 3-4%), was mottled, extremely firm and had acute capsular margins. Most areas of tissue were firmer than expected, but others were soft,



Liver, dog. Grossly, the liver is 50% normal weight, firm, and mottled. (Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, 1600 East Rollins Street, Columbia MO 65211)

when pressure was applied across a tissue slice. The kidneys were congested and swollen.

Laboratory results: ALT values were >2000; ALP, GGT and total bilirubin were also elevated.

The animal was serologically positive for antibody against both *Ehrlichia canis* and *Rickettsia rickettsia.* PCR tests did not detect adenoviral, influenza of leptospiral antigens. Positive results were obtained for parvovirus and distemper. Toxicologic testing failed to detect arsenic or aflatoxins.

Microscopic description: The liver is collapsed and architecturally only bile ducts and a few lipid-rich, degenerating, rounded hepatocytes remain associated with the sinusoids. Necrotic cells frequently have pyknotic nuclei and vacuolated cytoplasm containing bile pigment is common amongst them. Scattered mixed leukocytes remain in the sinusoids or in the adventitia of portal areas. Lymphocytes and plasma cells are particularly numerous in portal triads. Collecting veins and sinusoids focally contain thrombi.

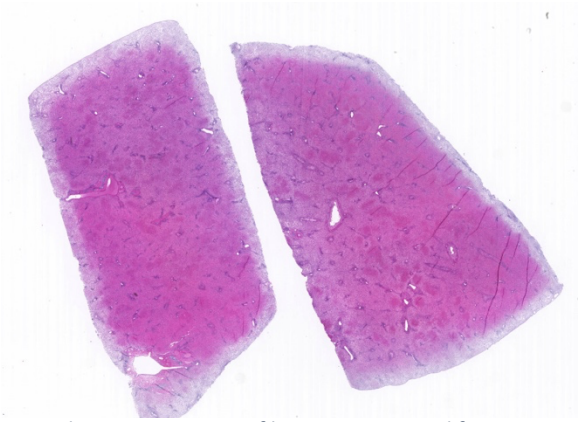
Contributor's morphologic diagnosis: Massive hepatic necrosis and loss, with periportal hepatitis

Contributor's comment:
Potentiated sulfonamides, combined with trimethoprim have become increasingly used to treat a variety of bacterial infections, especially those of the lung, skin and urinary tract in dogs. The two drugs are synergistic in inhibiting folate synthesis, inhibiting two steps of the pathway.

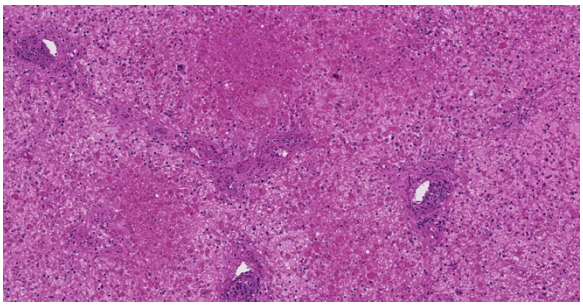
There are several manifestations of TMS toxicity in dogs, including anemia, non-septic arthritis, erythema multiforme-like skin syndrome and liver disease.[14,16] Hepatopathy is the most frequently lethal manifestation of toxicity. Cholestasis and/or massive or sub-massive hepatic necrosis are commonly found.[12,15] In some instances, lymphoplasmacytic infiltrates occur in lesions. Dogs can show multiple manifestations of toxicity. The drug dose typically is not excessive.

Illness commonly follows exposure by several days, so that the animal may not be currently taking the drug or be near the end of treatment. If surviving dogs are re-exposed to sulfa drug (with or without trimethoprim), recurrent disease is likely. It is thought that metabolic intermediates from drug detoxification produced by microsomal cytochrome P450 result in either a nitrosylated toxic metabolite or a hapten, perhaps one that binds to membranes and incites immune reaction that is lethal to the cell.

Onset of disease can occur near the end or after the antibiotic course, but disease extends beyond withdrawal of the drug. Some dogs that are affected have been given the drug previously. Clinicopathologic findings include increased alanine amino transferase, bilirubin, and alkaline phosphatase.[11]



Liver, dog. Two sections of liver are presented for examination. At subgross magnification, areas of hemorrhage are visible. (HE, 327X)



Liver, dog. There is necrosis and hemorrhage within centrilobular and midzonal areas of the hepatic lobule; portal hepatocytes are markedly vacuolated. (HE, 124X)

Breed predispositions occur but vary between studies.[4,5,14] At least one Saint Bernard has been reported with fatal liver disease, and Dobermans appear over-represented. The finding of related animals becoming ill in this case is striking.

The mechanism of toxicity is somewhat explored. Dogs do not have acetyl-methyl transferase activity, which is credited with producing the major non-toxic metabolite in people.[13] The alternative pathway is biotransformation by hydroxylation and then reduction to produce a nitrosylated reactive product. The lack of N-acylation occurs in all dogs, but Doberman dogs, including an affected dog, had cells with intrinsic *in vitro* toxicity at lower dose indicative of a second mechanism.[5]

A variety of other drugs can also cause hepatic toxicity, including anesthetics halothane and methoxyflurane, anticonvulsants and others. Consumption of natural toxins such as microcytin can also result in similar lesions.

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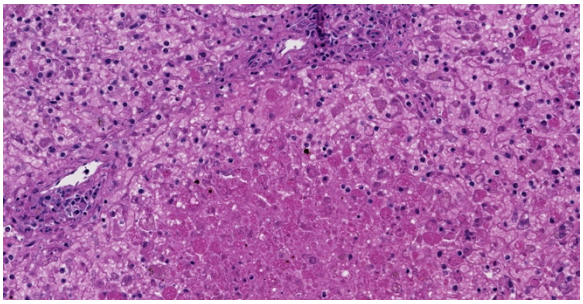
JPC diagnosis:
Liver, hepatocytes: Necrosis, massive, diffuse, with marked hemorrhage.

JPC comment:
Research on adverse reactions in humans to sulfa drugs reported from 1968 to 1988 illustrated several key points. The research centered on using sulfa drugs as a primary or adjunct therapy in the treatment of malaria, as the effectiveness of chloroquine waned. Reported reactions from sulfa drug therapy included blood dyscrasias (15%), skin lesions (45%), and liver related disease (7%). Drugs in the co-trimoxazole family accounted for 67% of all reported adverse reactions, which includes the trimethoprim/sulfamethoxazole family of drugs, still widely used in veterinary medicine. This category of sulfa drug was also responsible for 60% of the case fatalities in this reported period. There is also an association between longer sulfa drug half-life and increased case fatality rate.[1]

This case also provides an opportunity to review causes of acute massive hepatic necrosis in domestic species. Other causes of massive hepatic necrosis include other drugs, toxins that damage the hepatocyte cytoskeleton, the inhibition of RNA synthesis, iron toxicity, diet, and viruses.

- Carprofen in dogs and diazepam in cats has been reported to cause idiosyncratic diffuse and massive hepatic necrosis at therapeutic doses. Diclofenac causes similar idiosyncratic liver injury to humans.[6]
- *Microcystis aeruginosa*, a freshwater cyanobacteria, creates hepatotoxin microcystin-LR, which inhibits hepatocellular protein phosphatases 1 and 2A, disrupting the cytoskeleton.[3]
- *Amanita* sp. mushrooms create the toxin phalloidin, which binds to actin and disrupts the cytoskeleton.[3]



Liver, dog. There is loss of sinusoidal architecture in centrilobular and midzonal areas, necrosis of hepatocytes, hemorrhage and erythrophagocytosis by Kupffer cells. Periportal hepatocytes have numerous lipid vacuoles in their cytoplasm. (HE, 279X)

- Marine dinoflagellates create pectenotoxins, which directly damage the hepatocyte cytoskeleton.[3]
- Aflatoxin (**B_1**, B_2, G_1, and G_2) cause widespread hemorrhage and massive hepatic necrosis, especially in young animals.[6]
- *Amanita* sp. mushrooms elaborate α-, β-, γ-, and ε-amanitin toxin, which are potent inhibitors of RNA-polymerase, effectively halting RNA synthesis in hepatocytes.[6,8]
- Both iron-dextran intoxication of piglets, and ferrous fumarate in foals have been described as causes of massive hepatic necrosis.[2]
- Hepatosis dietetica in pigs is incompletely understood but is likely related to deficiency of vitamin E and/or selenium.[6]
- Theiler's serum hepatitis in horses may be caused by a parvovirus, though research continues to build upon the body of knowledge.[7]
- *Sulawesi tortoise adenovirus-1* (STAdv-1) was discovered to cause mortality in Sulawesi tortoises and has the capacity for interspecies transmission and infection.[9]
- *Rift Valley Fever virus* results in multifocal to massive hepatic necrosis in aborted lamb fetuses.[10]

Distinguishing between these causes can be difficult, and signalment and clinical history can provide the insight needed. This case highlights the need for additional information and analysis of lesions of other systems.

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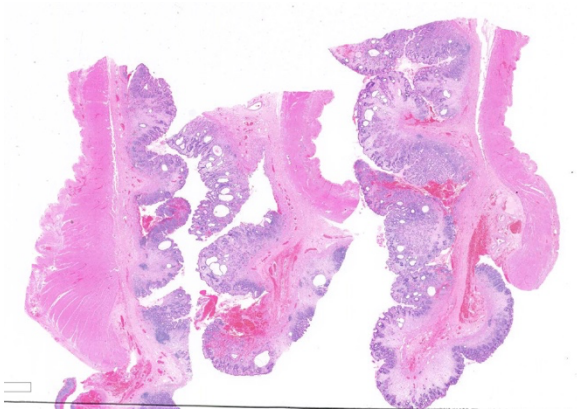


Stomach, dog. Three sections of stomach are presented for examination. At subgross magnification, the edematous mucosa is through into rugose folds. (HE, 7X)

CASE 2: 4884 A1 (4141105-00)

Signalment: Adult, neutered male, Border collie, *Canis lupus familiaris*

History: This case was referred to the hospital, where it presented with mild chronic hypoglycemia [blood glucose=2.8mmol/L (range: 3.6-7.0)] and high blood insulin [blood insulin= 72uIU/L (range: 5.0-40)]. Similar findings had been previously reported by the referring vet. This dog had one previous episode of seizure, which was suspected to be secondary to hypoglycemia. Ultrasonography and CT revealed a gastric fundic mass, without gastric lymphadenopathy. Cytology taken from the gastric fundus was non-conclusive. Finally, exploratory laparotomy was undertaken during which subtle thickening of the wall of the fundic stomach was noted. A transmural biopsy sample was taken during this surgery.

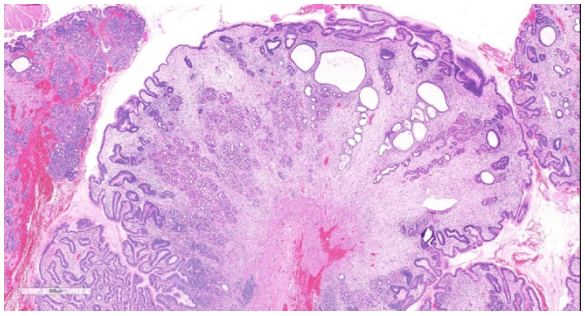
Gross Pathology: The sample submitted for histological examination was a 7cm x 7cm x 1.25cm transmural resection with diffuse, poorly defined, thickened and roughened mucosa.

Microscopic description: There is moderate to marked, diffuse thickening of the gastric mucosa, which forms sessile and polypoid expansions into the lumen. This is due to accumulation of large numbers of glands lined mostly by a cell thick layer of columnar epithelium with basophilic to clear cytoplasm (mucus containing cells), with occasional areas where cells pile up. Moderate numbers of these glands are markedly dilated and lined by attenuated epithelium. Most of the above-mentioned glands have basal, intraepithelial infiltration by small lymphocytes. The lamina propria in these areas is infiltrated diffusely by lymphocytes and plasma cells. At the base of areas of mucosal expansion there are pockets of fundic glandular tissue (preexisting), featuring occasional intraluminal necrotic cells, and lymphoid aggregates in the lamina propria (gut associated lymphoid tissue). Additionally, there are multifocal areas of extravascular accumulation of erythrocytes in the lamina propria (hemorrhage). There is no evidence of invasion of the submucosa or the basal lamina by the hyperplastic tissue described above.

Contributor's morphologic diagnosis: Marked, diffuse, mucosal hypertrophy with chronic gastritis, mucoid metaplasia, and glandular dilation – fundic stomach

Contributor's comment:
Gastric mucosal hypertrophy (GMH) is a canine disease that can be focal or diffuse and leads to mucosal proliferative changes. The terminology used to describe this disease can be somewhat confusing. As background, the concepts of hypertrophy and hyperplasia in both the macroscopic (gross) and microscopic settings are relevant in GMH (pathogenically and/or terminologically):[6]

- *Hypertrophy* (Greek etymology: hupér – over + trophia – nourishment). It describes the increase in volume of a



Stomach, dog. There is marked loss of gastric glands. More superficial glands are lined with poorly differentiated epithelium; glands deeper in the mucosa are typical, although atrophic. Numerous glands are markedly ectatic. (HE, 23X)

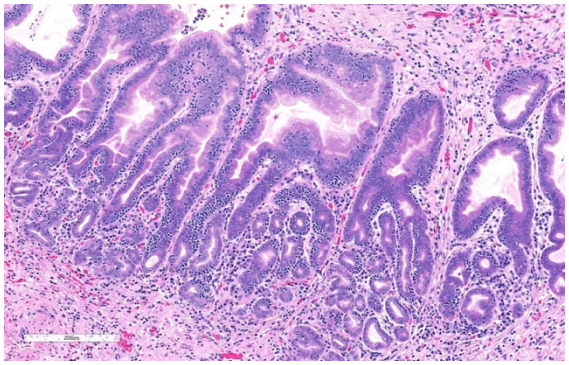
tissue or organ (gross) due to increase in parenchymal cell size (microscopic).

- *Hyperplasia* (Greek etymology: hupér – over + plasis – formation). It refers to increase in volume of a tissue (gross) due to increase in the number of parenchymal cells (microscopic)

With the above in mind, it is clear that when the term hypertrophy/hypertrophic is used in the description of GMH, it refers to its gross presentation of this disease. Microscopically, GMH features both hypertrophy, and hyperplasia despite its name.

GMH can be focal or diffuse:

- Focal GMH (F-GMH) is also known as giant hypertrophic pyloric gastropathy and may feature hypertrophic antritis (i.e. hypertrophy of the pyloric antrum).[4,9] It is idiopathic, relatively more prevalent than the diffuse form below, and it more frequently affects small canine breeds. In F-GMH, the proliferative lesion is located on the pyloric antrum, and consists of papillary proliferations into the lumen, which frequently lead to obstruction. Histologically, these proliferations consist of a mucosa thickened by increase in the amounts of surface epithelium foveolar tissue. Both of these are lined by epithelial cells with intracytoplasmic mucus (metaplasia). These are supported by a lamina propria that is either histologically normal, or contains plasma cells, lymphocytes, and eosinophils. It may be associated with local muscularis hypertrophy and/or mucosal erosion/ulceration.



Stomach dog. Glands within the superficial mucosa are lined by poorly differentiated basophilic mucosa and surrounded and infiltrated by numerous lymphocytes (HE, 175X)

- Diffuse GMH (D-GMH) can also be known as chronic hypertrophic gastritis or chronic giant hypertrophic gastropathy.[4,9] It is also idiopathic although an immune mediated pathogenesis is suspected. DS-GMH is relatively rarer than F-GMH and can affect a range of dog breeds. Breeds overrepresented include Basenji, beagle, boxer and bull terriers. It is associated with vomiting, diarrhea, weight loss, protein-losing gastropathy, and hypoproteinemia. Its presentation is similar to that described for human diseases with Ménétrier disease (more on this below). The gastric mucosa is diffusely thickened to form "cerebriform" folds that do not resolve upon stretching of the gastric wall. These may involve an area of 4–10cm only (which contrasts with the use of the term diffuse in the terminology above). Microscopically, there is hypertrophy and hyperplasia of the mucosa to form folds that may or may not contain submucosa and muscularis layers. The hyperplastic mucosa is composed of foveolar and glandular tissue, and there is progressive loss of parietal cells, that are replaced by variably differentiated mucus containing cells. A distinctive feature is the presence of inflammatory infiltration -and occasionally edema- of the lamina propria, which is mononuclear (i.e. lymphocytes and plasma cells with rarer macrophages). Partial gastrectomy can be curative.[10]

Overall, the histological presentation of the sample of stomach in this case is consistent with D-GMH, as this diagnosis is supported by the location (i.e. fundic stomach), gross presentation, and histological features. A striking feature of this presentation are the intraepithelial lymphocytes. More investigation would be required to ascertain their role (e.g. immunohistochemical characterization), but

these are suspected to be secondary to inflammation. One puzzling aspect of this presentation is the hypoglycemia, which is not a feature of the presentation of any of the diseases above described. It is therefore possible that this feature was not associated with the gastric lesion.

Parallelism between GMH (a.k.a. chronic hypertrophic gastritis or chronic giant hypertrophic gastropathy) and human Ménétrier disease is often drawn. Ménétrier disease is a hypertrophic gastropathy also characterized by "cerebriform" thickening of the gastric fundus/body mucosa secondary to excessive production of transforming growth factor alpha (TGF-alpha).[8] It affects adults (30-60y) most frequently, but a self-limiting pediatric form is reported (usually following respiratory infection). It is characterized by foveolar epithelial hyperplasia (with mucus containing cells), parietal and chief cell atrophy, and mild to moderate inflammation. Interestingly, striking intraepithelial lymphocytosis is present in some cases, in consistency with the canine D-GMH case presented here. Ménétrier disease in adult humans is associated with increased risk of gastric adenocarcinoma.

Canine gastric adenocarcinoma (GA) is a differential diagnosis to consider for this presentation.[9] There are several features of GA that contrast with the presentation
noted in this case, however. Grossly, GAs cause loss of the rugal aspect of the mucosal surface, may be ulcerated (>50% of cases), and are frequently located in the pylorus/antrum. Histologically, they frequently have severe desmoplasia, and are heavily infiltrative, with frequent invasion of the lamina propria progressing into the muscularis and serosa to form a transmural lesion. Also, lymphatic invasion and metastasis to local lymph nodes and other distant organs (especially lung, liver and adrenal glands) are common occurrences in GA. None of these were noted in this case.

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https://www.ed.ac.uk/vet/services/easter-bush-pathology

JPC diagnosis:
Stomach: Gastric hypertrophy, chronic, diffuse, severe, with mucoid metaplasia, marked glandular atrophy and loss, and moderate lymphocytic inflammation.

JPC comment:
First described in 1888 by French pathologist Pierre Ménétrier, this disease was first characterized by "sheet-like polyadenomas" of the gastric mucosa affecting the proximal portion of the stomach (body and fundus) but sparing the distal stomach (antrum). Many of his findings remain characteristic for the eponymous Ménétrier's disease.[3]

The contributor provided a good review of this entity. In humans, Ménétrier's disease is also known as hyperplastic gastropathy, or giant rugal hypertrophy. A familial link has been rarely documented but shows no regular association with other diseases. Interestingly, cytomegalovirus is present in approximately 70% of cases in affected children, and *Helicobacter pylori* is detected in almost 90% of adults affected.[2]

Giant hypertrophic gastritis, or Ménétrier-like disease, has been reported in an Old English sheepdog, a Jack Russell terrier, and a Boxer. A Ménétrier-like disease has been reported in association with gastric carcinoma in a West Highland white terrier, and gastric adenocarcinoma in Cairn terrier littermates. The contributor previously mentioned the breed specific form of gastric hypertrophy in Baseneji, though that histologic appearance differs from Ménétrier-like disease exhibiting only infrequent glandular cysts and a concurrent lymphoplasmacytic gastroenteritis. A recent report of Ménétrier-like disease in a domestic shorthair cat found a predominant mucosal hyperplasia, dilated gastric glands, a mild interstitial fibrosis and inflammatory infiltrates.[1]

A hypertrophic gastropathy similar to Ménétrier's disease has been reported in

transgenic mice. A line of mice was created that overexpressed transforming growth factor a (TGF-α) in the stomach. The mice exhibited dramatic structural and functional lesions of the glandular stomach, including severe adenomatous hyperplasia and a lack of detectable gastric acid. It remains possible that TGF-α plays an important role in the development of pathology similar to Ménétrier's disease.[7]

Important differentials to consider for diffuse hypertrophic gastropathy include Ménétrier's disease, hypertrophic lymphocytic gastritis, hypertrophic hypersecretory gastropathy, and Zollinger-Ellison syndrome. Hypertrophic lymphocytic gastritis can be differentiated by diffuse and severe inflammation with numerous intraepithelial lymphocytes. Hypertrophic hypersecretory gastropathy differs from Ménétrier's disease by exhibiting hyperplasia of both the foveolar epithelium and oxyntic glands, with cystic dilatation of the gastric glands possible. Zollinger-Ellison syndrome is characterized by a lack of foveolar hyperplasia, but with ectopic gastrin secretion (gastrinoma) with increased gastric acid secretion, intractable peptic ulcers, diffusely thickened gastric folds, and diffuse parietal cell hyperplasia and hypertrophy.[5]

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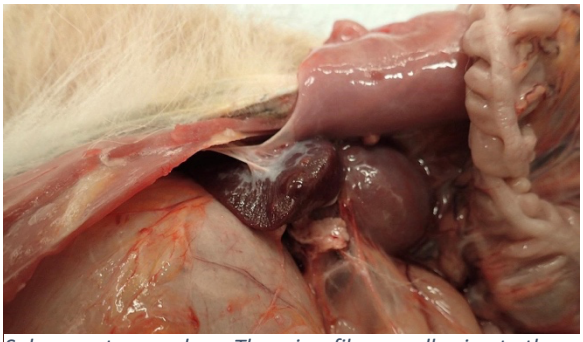


Spleen, patas monkey: There is a fibrous adhesion to the body wall, and several nodules raised above the capsular surface. (Photo courtesy of: Integrated Research Facility, Division of Clinical Research, 8200 Research Plaza, Fort Detrick, MD 21702, https://www.niaid.nih.gov/about/integrated-research-facility)

CASE 3: 2019 Case 2 (4135953-00)

Signalment: Intact, female, 24-year-old, patas monkey (*Erythrocebus patas*)
History: This monkey presented with bloat prior to humane euthanasia.

Gross Pathology:
Spleen:

1. Chronic, focal, moderate fibrous capsular adhesion to the body wall
2. Multifocal, firm, 0.3-0.8 cm, dome-shaped, raised nodules

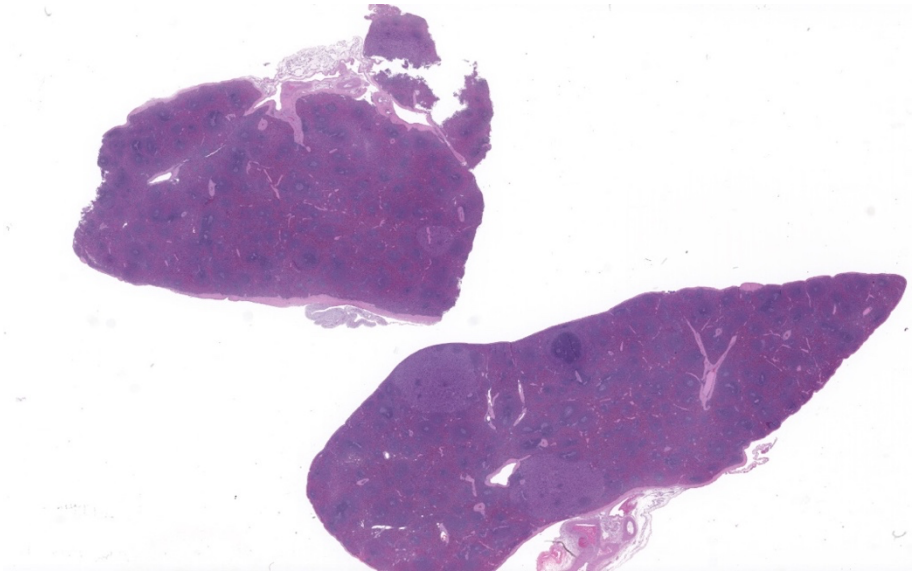
Laboratory results:
See table.

Assay	Units	Value	Min	Max		Assay	Units	Value	Reference Range
WBC	10^3/uL	**5.91**	2.14	5.91		**Glucose**	mg/dL	**291**	60-120
RBC	10^6/uL	**6.72**	5.3	6.95		**Blood Urea Nitrogen**	mg/dL	**40**	15-43
HGB	g/dL	**15.7**	12.6	17.5		**Creatinine**	mg/dL	**1.4**	0.7-1.4
HCT	%	**50.3**	39.4	53.7		**Calcium**	mg/dL	**9.5**	8.2-10
MCV	femto	**74.9**	73.4	82.6		**Albumin**	g/dL	**2.5**	2.0-4.1
MCH	pico	**23.4**	23.4	26.8		**Total Protein**	g/dL	**5.4**	4.8-6.7
MCHC	g/dL	**31.2**	31.2	33.7		**Alanine Aminotransferase**	uL	**40**	38-119
PLT	10^3/uL	**383**	223	383		**Aspartate Aminotransferase**	uL	**73**	37-91
Neut#	10^3/uL	**4.59**	0.46	4.592		**Alkaline Phosphatase**	uL	**153**	62-214
Lymph#	10^3/uL	**0.68**	0.68	2.211		**Total Bilirubin**	mg/dL	**0.5**	0.4-0.9
Mono#	10^3/uL	**0.52**	0.091	0.52		**Gamma-glutamyl transpeptidase**	uL	**36**	24-125
EO#	10^3/uL	**0**	0	0.241		**Amylase**	uL	**766**	333-575
Baso#	10^3/uL	**0.12**	0.039	0.21					
Ret#	10^6/uL	**0.0383**	0.034	0.0731					

CBC and serum chemistry acquired the day ***prior to*** bloating and euthanasia.
CBC reference ranges were acquired from 16 patas monkeys.
Serum chemistry reference ranges were acquired from 23 patas monkeys.
The clinical significance of the hyperglycemia and increase in amylase in this patas monkey are not known. A urinalysis was not performed.

Microscopic description:
Spleen (2 sections): Multifocally, the splenic parenchyma contains several nodular aggregates of polygonal to elongated mesenchymal cells forming variably sized, vascular channels separated by scant to moderate amounts of eosinophilic stroma. The cells lining the vascular channels are plump, contain open-faced, oval to elongated nuclei containing one to four nucleoli and scant eosinophilic cytoplasm. Anisocytosis and anisokaryosis are minimal. The vascular channels formed by this mesenchymal cell population are minimally to widely separated by a bland, eosinophilic stroma that largely replaces the pre-existing red pulp in locally extensive areas. In areas where the stroma accompanying these vascular channels is abundant, these vascular channels form distinct, unencapsulated nodules. Small numbers of hemosiderophages and neutrophils infiltrate these nodules multifocally. The splenic hilar connective tissue is mildly edematous diffusely and infiltrated by small numbers of neutrophils and macrophages. A densely collagenous, well-vascularized fibrous adhesion is attached to the capsule of the spleen. The mesothelium lining the splenic capsule is plump diffusely (reactive) and within one focal area, the mesothelium is 8 cells in thickness.

Contributor's morphologic diagnosis:



Spleen, patas monkey: Subgross magnification of the submitted sections of spleen demonstrates several discrete parenchymal nodules, one elevating the capsule in the lower section. (HE, 7X)

Spleen:

1. Sclerosing angiomatoid nodular transformation (SANT, presumptive)
2. Chronic, locally extensive, moderate capsular fibrosis (adhesion)

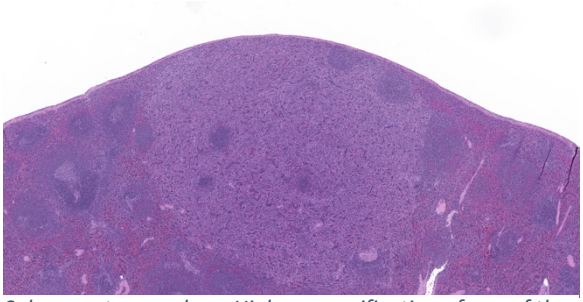
Contributor's comment:

First reported as "cord capillary hemangioma" in 1993[8,9], less than 150 SANT cases have been reported in the literature and the pathogenesis is poorly understood. The actual number of cases in the literature is unknown as the same lesion has been referred to by different authors as a hamartoma, hemangioma, and multinodular hemangioma. As the lesion is considered benign, differentiating between these morphologic diagnoses has questionable clinical relevance.[1,6,8] SANTs are thought to be a reactive, post-inflammatory or post-thromboembolic lesion.[2] Martel *et al*. describes the lesion as, "altered red pulp tissue that had been entrapped by a nonneoplastic stromal proliferative process."[11]

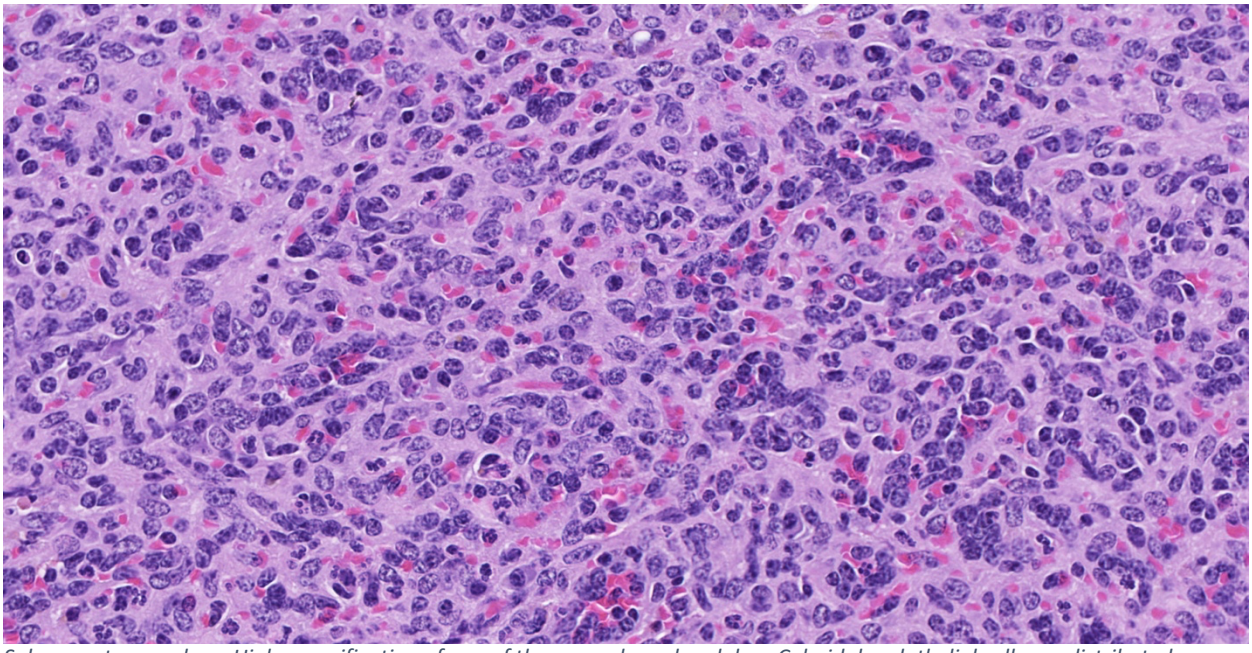
In humans, the diagnosis is most often reported in middle-aged women, discovered as an incidental finding or as the result of abdominal pain and/or splenomegaly.[11] Diagnostic features of SANT are their angiomatoid sclerotic nodular histopathologic appearance and mixed vascular component. Definitive diagnosis requires immunohistochemistry.[2,16]

Immunohistochemically, SANT lesions recapitulate the normal composition of the red pulp and contain vessels with three immunophenotypically distinct phenotypes: 1) capillaries that are CD34 positive/CD31 positive and reported to be either CD8 positive or negative (most commonly reported to be CD8 negative), 2) venules that are CD34 negative/CD31 positive/CD8 negative, and 3) red pulp sinusoids that are CD34 negative/CD31 positive/and CD8 positive.[2,12,15,16] Vessels within the lesion do not stain for D2-40 confirming they are not lymphatic in origin.[8,13] Variable positivity has been reported in SANT for the TNF-receptor transmembrane glycoprotein CD30[2,16] but the significance of the presence or absence of this marker is not known. Most plasma cells in these lesions are IgG4 positive.[2] In human cases, the cells composing the stroma stain variably for smooth muscle actin (pericytes), CD31 (endothelial cells), and CD68 (histiocytes).[11]

Differential diagnoses for SANT include littoral cell angioma, splenic hamartoma, splenic hemangioma, splenic hemangioendothelioma, and splenic angiosarcoma. The lesion in the spleen of this patas monkey has no dense foci of inflammation consistent with an inflammatory



Spleen, patas monkey: Higher magnification of one of the parenchymal nodules, this one elevating the overlying capsule. Several nodules of white pulp remain within the boundary of the nodule. (HE, 30X)



Spleen, patas monkey: High magnification of one of the parenchymal nodules. Cuboidal endothelial cells are distributed throughout, some forming blood-filled vascular spaces. Endothelial cells are separated by a moderate collagenous matrix, often with extravasated erythrocytes. (HE, 400X)

pseudotumor (IPT) or dense, avascular scarring (post-traumatic fibrosis) but these should be included in the differential diagnosis of multinodular splenic lesions with an extensive inflammatory or fibrous component.

SANTs lack the proliferative activity of and cytologic atypia seen in angiosarcomas. Angiosarcomas lack a nodular growth pattern, are invasive, and are positive for CD68 and CD8.[4,15] Hemangioendotheliomas (spindle, polymorphous, and epithelioid types) are considered intermediate in biological behavior between hemangioma and angiosarcoma have mild to moderate atypia and generally a low mitotic index[14] but share some histopathologic features with SANT and this diagnosis is favored by some for SANT lesions. Others favor a diagnosis of splenic hamartoma of the red pulp that has undergone sclerosis.[11,13] Hemangioendotheliomas, in addition to having greater cytologic atypia and mitotic activity than SANT, are variably immunoreactive for CD34 and cytokeratin, and are CD21, CD8, and CD68 negative. The vessels in splenic hamartomas are composed of only sinusoids (factor VIII positive/CD31 positive/CD8 positive/type IV collagen positive/CD21 negative/CD68 negative).[13] Both hemangioendotheliomas and splenic hamartomas lack the multinodular growth pattern seen in SANTs. Littoral cell (sinusoidal lining cell) angioma and splenic hemangiomas also lack the multinodular, sclerotic architectural features of SANT and are also composed of a single type of blood vessel. Littoral cell angiomas are typically CD31 positive/CD34 negative/CD8 negative/CD68 positive/CD21 positive. Splenic hemangiomas are CD31 positive/CD34 positive/CD8 negative/CD21 negative/CD68 negative. Treatment for this presumed nonneoplastic lesion, is splenectomy.[13] Immunohistochemistry for CD34, CD31, CD8, CD21, D2-40, factor VIII, and CD68 are pending in this case to confirm the presumptive diagnosis of SANT.

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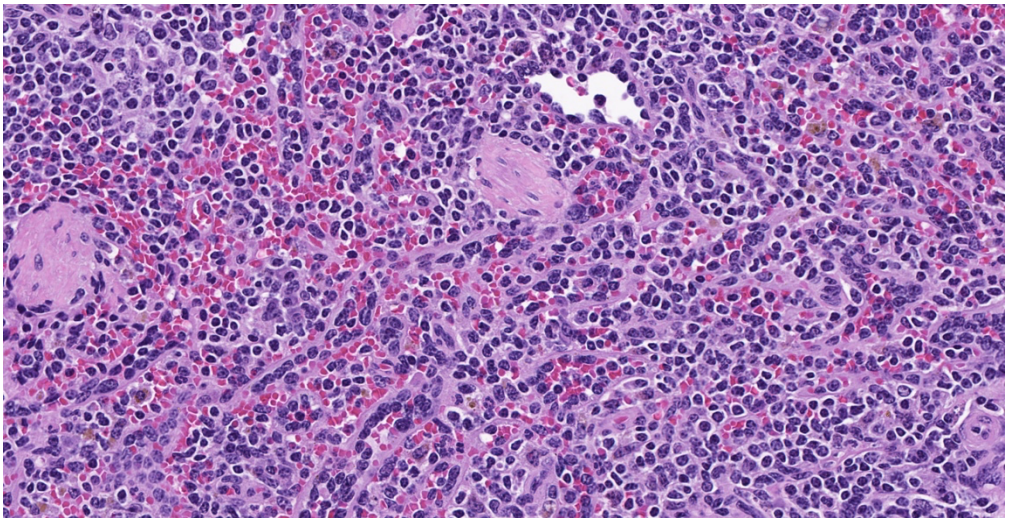
JPC diagnosis:



Spleen, patas monkey: Similar vessels lined by cuboidal epithelium are scattered through the red pulp. (HE, 400X)

Spleen: Vascular proliferation, diffuse, moderate, with endothelial hypertrophy and random nodular sclerosis.

JPC comment:
This succinct summary outlines the highlights of what has been learned since this was first described in 2004, and prior to that was likely diagnosed as other entities. One point that bears discussion is the nature of IgG4-related disease.

All plasma cells first secrete IgM until they receive stimulation to undergo isotype switching and produce IgA, IgE, or IgG antibodies. There are four subclasses of IgG, named in the both the order of discovery and concentrations found in circulation, from IgG1 to IgG4. While IgG4 shares more than 95% sequence homology in the constant domain with the other IgG isotypes, it differs significantly in a few amino acids in the CH2 domain. This leaves it with little affinity for complement C1q binding and for Fc receptor binding. The lack of a proline in the hinge region makes its inter-heavy chain disulfide bonds more prone to reduction, allowing heavy chains to swap between IgG4 antibodies. These are sometimes called 'half-antibodies' and allows them to function as 'bi-specific', but functionally monovalent for a given antigen. One disease involving IgG4 that is well understood is pemphigus complex, where the IgG4 antibody directly attacks desmoglein (or desmocollin) to affect pathologic changes in the epidermis.[12]

There is increased research interest regarding this process as IgG4 production and IgG4-positive plasma cells and histologic appearance are common threads for a variety of inflammatory diseases, including sclerosing forms of sialadenitis, pancreatitis, cholangitis, aortitis, thyroiditis, and retroperitoneal fibrosis in humans. In SANT, IgG4-positive plasma cells are a consistent finding, but whether this entity will be classified as an IgG4-related disease is still to be determined.[3,7,12]

SANT is a uniquely splenic lesion, though histologically similar entities exist outside of the spleen. One condition most often found in lymph nodes is called nodal angiomatosis and is a subtype of vascular transformation of sinuses with a more cellular character. While perhaps histologically similar, despite different tissues, they each have different IHC morphologies, with nodal angiomatosis typically demonstrating positive immunoreactivity to desmin, smooth muscle actin, and vimentin. Conversely, the spindloid cells of SANT are negative for smooth muscle actin.[5]

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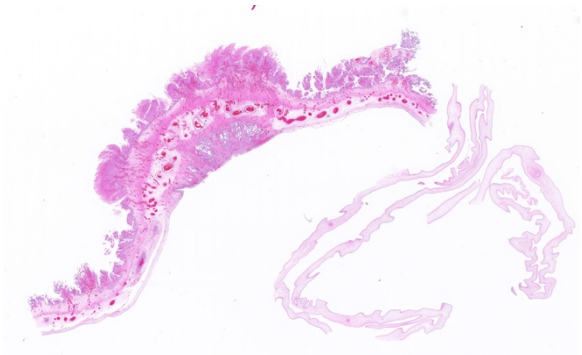
CASE 4: OSU 2013 2 (4041565-00)

Signalment: 1-day-old, male, Miniature horse (*Equus ferus caballus*)

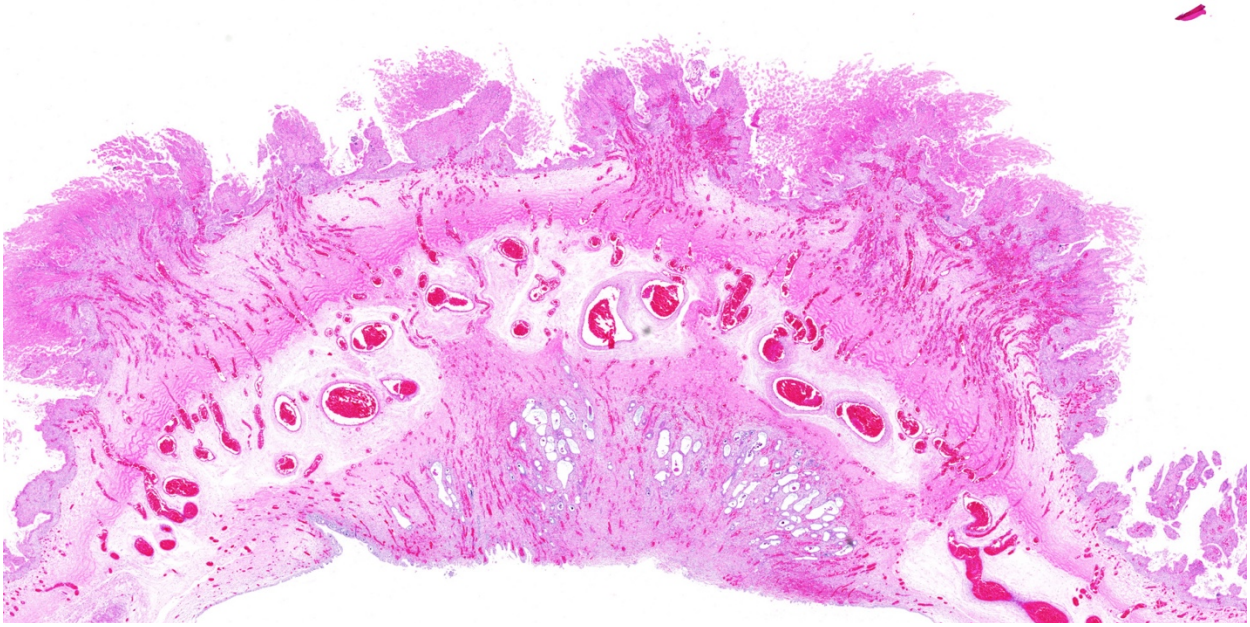
History: A dead, premature Miniature horse colt was delivered on 2/14/13 by a mare that presented with dystocia. The placenta passed approximately 10 minutes post-partum and while it appeared grossly normal, it weighed 6.2 pounds (increased). The mare's due date was 3/8/13. The colt and placenta presented for a complete necropsy on 2/15/13.

Gross Pathology: The body condition score was 1.5/5 and the animal presented in good post-mortem condition. A mild amount of yellow-brown, soft material was present on the tail, perineum, and all four limbs.

The endocardium of the heart contained two areas of red discoloration that extended 1-2 mm within the myocardium. The first area measured 5 x 5 mm and was located just ventral to the mitral



Placenta, horse. A section of placenta is presented for examination. At subgross examination, there is a nodule extending from the allantoic surface (arrow). (HE, 5X)



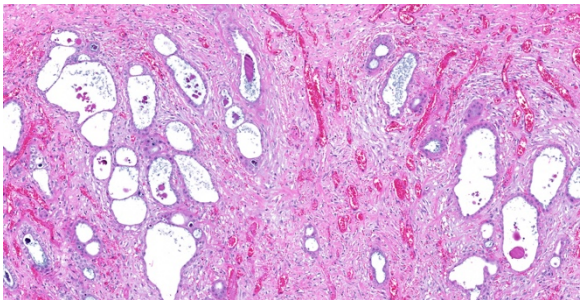
Placenta, horse. Higher magnification of the section of placenta containing the allantoic nodule (at bottom). There is necrosis of the chorionic villi opposing the nodule and marked proliferation of capillaries supplying those villi. There is marked congestion of the chorioallantoic vessels, and mild edema of the surrounding stroma. (HE, 14X)

valve; the other are measured 5 x 8 mm and was present within the papillary muscle. Bilaterally, both adrenal glands were characterized by markedly thin cortices, with a cortical to medullary ratio of 1:8. The cortical surface contained hundreds of pinpoint, red foci.

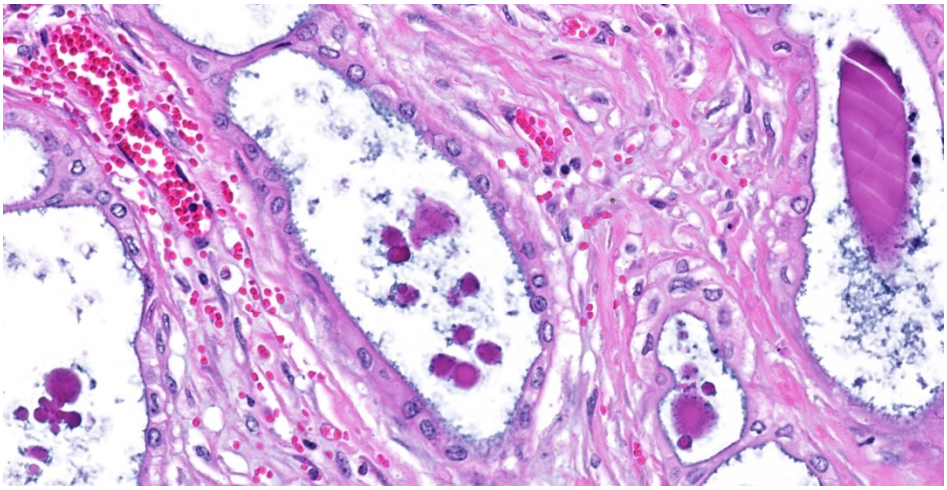
Both tips of the uterine horns contained dozens of raised, firm, tan irregular tissue that was moderately well-demarcated. The amniotic sac contained hundreds of raised, white, pinpoint, soft foci (amniotic plaques).

Microscopic description:
Placenta: A focal area of round epithelial lined structures is present within the fibrous stroma of the chorioallantois that forms a discrete nodule. The allantoic epithelium is stratified with piling of nuclei. Within the epithelium, dilated cystic structures lined by cuboidal epithelium with round nuclei. The cysts are filled with homogenous eosinophilic material. Similar dilated epithelial lined cystic structures are observed within the allantoic stroma of various sizes. Fibrosis, neovascularization and hemorrhage lie between the cysts in the chorioallantoic membrane. Edema is present below the chorionic villi, within the connective tissue. The chorionic villi demonstrate multiple foci of karyorrhexis and pyknotic epithelial nuclei, indicative of necrosis. Several regions of the villi are blunted. Few arteries and arterioles have clear cytoplasmic vacuoles within the tunica media and endothelium, interpreted as hydropic degeneration.

Contributor's morphologic diagnosis:
Marked multifocal to coalescing chronic allantoic epithelial fibroadenomatous hyperplasia with edema and neovascularization. Mild to moderate multifocal acute chorionic villus coagulation necrosis with villus atrophy.



Placenta, horse. There are dilated tortuous glands within the chorioallantoic membrane, which are separated by highly vascular fibrous tissue. There are variable amounts of secretory material within the glands. (HE, 181X)



Placenta, horse. Higher magnification of the glands, which are lined by cuboidal epithelium which is similar to allantoic epithelium. Condensed secretory material within their lumina form corpora amylaceae. (HE, 400X)

Contributor's comment:
Adenomatous hyperplasia of the allantoic epithelium is categorized in three stages. Stage 1 includes hypertrophic and hyperplastic allantoic epithelium with intraepithelial glands and intracytoplasmic vacuoles. Stage 2 includes the presence of glands within a thickened and reactive allantoic stroma. Stage 3 includes numerous glands of various sizes within the stroma forming a nodule.[6] This is a case of stage 3 fibroadenomatous hyperplasia of the allantoic epithelium. Adenomatous hyperplasia has been associated with other chronic placental lesions including nocardia, fungi, *Streptococcus spp., Pseudomonas spp.*, *Staphylococcus spp.*, and coliform (predominately *Escherichia coli)* placentitis.[6] However, these lesions are characterized by focal chorionic villar necrosis, and inflammatory cells within the mass and chorionic epithelium. Acute placentitis has not been associated with adenomatous hyperplasia suggesting that it may be a reactive lesion due to a chronic irritant.[6]

A study reviewing cases of placental leptospirosis reported 95% of affected placentas demonstrating allantochorionic lesions, the most frequent being adenomatous hyperplasia of allantoic epithelium. Also commonly observed was leukocyte infiltrates within the chorionic villi and stroma, vasculitis, and chorionic villar necrosis, atrophy, and mineralization.[7]

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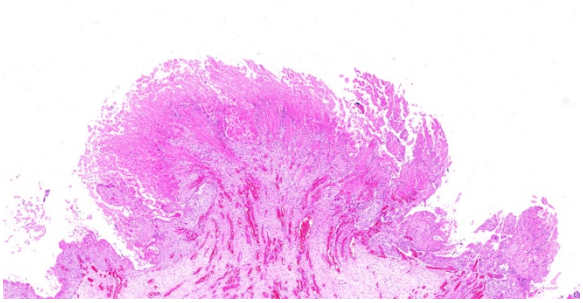
JPC diagnosis:
Placenta, allantois: Hyperplasia, adenomatous, focally extensive, moderate, with fibrosis, neovascularization, and coagulation necrosis of opposing chorionic villi.

JPC comment:
The condition was first reported in 1988 as adenomatous dysplasia of the equine allantois, but similar lesions have been reported near the umbilicus on the allantoic surface of placentas in



Placenta, horse. The choronic villi on the opposing face of the chorioallantoic membrane demonstrate partial to full-thickness coagulative necrosis. There is marked hyperplasia of capillaries supplying these distressed segments of the chorion. (HE, 36X)

elephants and rhinoceroses. These lesions have never been reported in other animals or humans.[5]

Adentomatous hyperplasia or dysplasia of the equine allantois is currently considered by most authors to occur secondary to chronic placental pathology of other causes.[1,3,8] The grading scale of lesions, which can be focal or multifocal, was described above. These lesions likely do not directly affect placental function directly. In a case series documenting 8 cases of cystic adenomatous hyperplasia of the allantois, many mares had normal foals prior to an abortion, and at least 3 went on to have subsequent normal foals.[8]

It is currently not known whether the criteria for the staging occur in a chronologic fashion, or different histologic features manifest independently. The degree, or stage, of disease does not appear to correlate with severity of any symptoms, extent of inflammation, or a particular pathogen.[3]

While we cannot know the underlying prevalence in the spectrum of healthy births, there is a high prevalence of abortion and bacterial placentitis associated with this condition. Because the histologic changes do not correlate with an acute placentitis, it has been proposed that this condition may predispose the mare to developing a secondary bacterial placentitis.[1,3]

An important differential for this entity is amnion nodosum, which is characterized by the presence of numerous small papules or granular amorphous nodules on the amnionic surface. A report of amnion nodosum was reported in a Belgian draught horse in 2011 and, similar to many horses affected by adenomatous dysplasia of the allantois, the mare and foal continued to be healthy with no adverse effects noted.[2,4]

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