

Veterinary Diagnostic Laboratory

UNIVERSITY OF MINNESOTA

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Veterinary Diagnostic Laboratory
College of Veterinary Medicine
1333 Gortner Avenue
St. Paul, MN 55108

Diagnostic Report

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Accession Number: D15-025835

Owner: WILDLIFE SCIENCE CENTER 851 WOLF
5463 W BROADWAY
COLUMBUS, MN 55025

Received Date: 05/13/2015

Site:

Reference:

Species: Miscellaneous Mammals

Premises ID:

Breed: Wolf

Submitted by: Credit Card Paying Client
Veterinary Diagnostic Laboratory
1333 Gortner Avenue
St. Paul, MN 55108
US

Age: 13 Years **Gender:** Female

Weight:

Status: Final

Preliminary: 05/21/2015 10:37:00

Final: 05/26/2015 11:45:00

History:

This wolf (13 year old, female spayed, Mexican Gray Wolf, "851") had a history of labored breathing and blood in the abdomen. The wolf was euthanized on 5/12/15. The necropsy was performed on 5/13/15 by Rachel Rice, Jennifer Vogel, Alyssa Rudloff, and AlexAnne Weinzierl (veterinary students) under the supervision of Dr. Armien and Dr. Sturos.

Specimen:

The whole body of a gray and tan aged female Mexican Gray Wolf was submitted in a state of fair postmortem preservation. Microchip identification was not detected using a universal microchip scanner.

Necropsy:

Body Condition Score : This 22.2 kg wolf had a body condition score of 2/5 (1 = emaciated and 5 = obese). The wolf was thin and had minimal subcutaneous and intra-abdominal adipose tissue.

General Findings : There were many brown dog ticks in various states of engorgement on the wolf. Brown liquid feces was oozing from the rectum as well as on the surrounding hair. The abdomen was distended with gas. The oral mucous membranes were pale to white.

Body cavities : The thoracic cavity contained 2,550 mL of hemorrhagic fluid.

Integumentary system : The distal pinna of the right ear was missing with an irregular scarred border.

Muscular system : There were no significant macroscopic lesions.

Skeletal system : There was severe white bony proliferation on the ventral vertebral bodies which bridged between adjacent vertebral bodies ventrally in the thoracic vertebrae from T4-T9.

Respiratory system : The lungs were mottled pink and red, moderately collapsed, and rubbery.

Cardiovascular system : There was 1000 mL hemorrhagic fluid within the pericardial sac. There were numerous raised, 1-2 mm diameter, tan to brown projections from the visceral pericardium and epicardium, which were composed of gritty granular material. The dorsal pericardium near the aortic root and the heart base epicardium were most severely affected, with small numbers of similar appearing projections overlying coronary vessels and occasionally surrounded by hemorrhage. There was moderate extensive subendocardial fibrosis of the left atrium. There was severe nodular thickening of the mitral and tricuspid valve leaflets and enlargement of the tendinae cordae. There was enlargement and fibrosis of the semilunar leaflets of the aortic valve. The sinus of Valsalva was dilated to 1.5 cm in diameter. The muscular septum was replaced by one centimeter in diameter fibrous membrane in the subaortic position. The heart weighed 252 grams (1.1% of body weight). The right ventricular free wall, left ventricular free wall, and interventricular septum measured 5, 8, and 12 mm, respectively. There was a extensive area of fibrosis affecting 40% of the right papillary muscle of the left ventricle. The circumference of the aortic artery was 6.5 cm. The circumference of the pulmonary trunk was 5 cm. The circumference of the base of the heart was 22.3 cm.

Alimentary system : The liver weighed 658 grams (2.9% of body weight). There was a poorly demarcated, soft, 3 cm diameter, dark red and black speckled raised irregularly ovoid mass in the parenchyma of the right medial lobe. The liver was crepitant on palpation. The teeth showed severe generalized surface wear. The stomach contained yellow/brown liquid.

Urinary system : There were no significant macroscopic lesions.

Endocrine system : There were no significant macroscopic lesions.

Reproductive system : The wolf was a spayed female.

Hemolymphatic system : There were no significant macroscopic lesions.

Nervous system : There were no significant macroscopic lesions in the eyes or brain.

Histopathology:

Slide A,B: Pericardium – The parietal pericardium is variably expanded by plaques composed of predominantly large round to ovoid to occasionally elongated cells which are moderately to markedly vacuolated and have a round to oval nucleus with finely stippled chromatin and 1-2 nucleoli (interpreted to be macrophages, or less likely, mesothelial cells) and few lymphocytes and plasma cells, supported by poorly organized hyalinized to fibrovascular stroma. Admixed with the macrophages/mesothelial cells, lymphocytes and plasma cells are small numbers of cells which contain large amounts of multilocular or unilocular clear cytoplasmic material (fat) and have flattened, peripheral nuclei (interpreted as adipocytes). The plaques do not extend into or through the fibrous pericardium. In areas there are papillary projections arising from the parietal pericardium which are composed of central hyalinized collagenous stalks and fronds lined by plump hyperplastic mesothelial cells and small numbers of macrophages, lymphocytes and plasma cells within the stroma; however, often the lining epithelium is sloughed. There are aggregates of amorphous eosinophilic material (fibrin and cellular debris) and occasionally yellow granular material (interpreted as hematoidin) adhered to the surface or trapped within fronds of the papillary projections.

Atrioventricular valve – The spongiosa of the valve leaflet is expanded by myxoid stroma and fibroblasts and the fibrosa is disrupted and disorganized in several areas.

Slide C: Atrium – The epicardium multifocally thickened with plaques and papillary projections with similar architecture and cytologic features to the changes described in the pericardium in slides A and B. In the subepicardial myocardial interstitium there are small numbers of lymphocytes, macrophages and neutrophils. Several medium caliber subepicardial arteries are infiltrated by small numbers of macrophages, lymphocytes, and neutrophils within the medial and adventitial layers and linear to circumferential bands of amorphous eosinophilic material (fibrinoid necrosis).

Slide D, E: Right ventricle – Similar to the pericardium and atrium previously described, the mesothelial surface of the epicardium is thickened by plaques and papillary projections with occasional infiltration of the subepicardial myocardium by lymphocytes, macrophages and plasma cells. Similar the atrium there are several vessels with fibrinoid necrosis of the media and adventitia.

Slide F: Left ventricle – Within the papillary muscle there are arborizing areas of cardiomyocyte loss and replacement fibrosis affecting 35% of the area of the papillary muscle. Epicardial changes are as previously described.

Slide G: Junction of left ventricle and interventricular septum – There is extensive fibrinoid necrosis of several small and medium caliber subepicardial arteries. Other epicardial changes are similar to those previously described.

Slide H, I: Lung – There is diffuse congestion of the alveolar capillaries. The alveolar lumina contain homogenous pale eosinophilic material (edema) or are partially or completely collapsed (atelectasis). There is a small focus of subpleural septal fibrosis, type II pneumocyte hyperplasia and increased alveolar macrophages within the alveoli of one section of lung. There is moderate postmortem autolysis.

Slide J: Kidney – There is mild to moderate thickening of the mesangial interstitium. There is marked postmortem autolysis.

Thyroid – There are no significant microscopic lesions. There is moderate postmortem autolysis.

Slide K: Spleen – There are no significant microscopic lesions. There is moderate postmortem autolysis.

Liver – There is marked postmortem autolysis which precludes histopathologic interpretation.

Slide L: Adrenal gland – There are nodular areas of hyperplastic cortical epithelial cells. There is moderate postmortem autolysis.

Slide M: Stomach, small intestine – There are no significant microscopic lesions. There is marked postmortem autolysis.

Special Stains:

Gram, GMS, and Fite's modified acid-fast stains were applied to serial sections of block A and block G. Small numbers of individualized rod bacteria (both Gram positive and Gram negative) were present in the superficial pericardial debris in block A but not within the deeper tissues. Infectious organisms were not present within block G.

Immunohistochemistry:

Vimentin, broad-spectrum cytokeratin, calretinin, CD18 and Mac387 immunohistochemistry were performed on serial sections of block A. *Borrelia* immunohistochemistry was performed on block G. The round to ovoid cells comprising the thickened plaque-like areas in the pericardium were a mixed population of both cytokeratin positive, CD18 negative and cytokeratin negative, CD18 positive. The superficial layer of

mesothelium (vimentin and cytokeratin positive cell population) that was expected to be present lining the pericardium was frequently absent in the examined sections (interpreted as sloughed due to postmortem autolysis). Mac387 and *Borrelia* antibodies exhibited non-specific background binding and were non-diagnostic. There was no immunoreactivity for calretinin within the examined section.

Bacteriology:

A swab of the pericardial and epicardial material was submitted for aerobic and anaerobic cultures. A mixed culture (3+) of *Klebsiella* species and non-hemolytic *Escherichia coli* were isolated on aerobic culture. Multiple species were isolated on anaerobic culture: *Clostridium perfringens* (2+), *Bacteriodes* species, *Clostridium sordellii*, *Clostridium* species, and *Peptostreptococcus* species.

Diagnosis:

1. Pericardium and epicardium –
 - a. pericarditis and epicarditis, lymphohistiocytic and proliferative, diffuse, moderate to focally marked, chronic
 - b. hemopericardium, marked, chronic
2. Heart –
 - a. myocardial atrophy, loss and replacement fibrosis (infarction), locally extensive (left ventricular papillary muscle), marked, chronic
 - b. atrioventricular valves (bilateral) and aortic valves, myxomatous valvular degeneration (endocardiosis), marked, chronic with left atrial subendocardial fibrosis
 - c. fibrinoid arteritis, multifocal – subepicardial, moderate to focally marked, chronic
 - d. interventricular septum, sub-aortic membranous septum, focal, moderate, chronic
 - e. Sinsus of Valsalva - aneurism
3. Thoracic cavity – hemorrhagic hydrothorax, marked, subacute to chronic
4. Lung – atelectasis, diffuse, moderate, subacute to chronic
5. Vertebral column (thoracic vertebrae) – spondylosis, multifocal, marked, chronic (gross diagnosis)

Comments:

This wolf was euthanized. The reported clinical sign of labored breathing was due to the large amount of blood in the pericardial sac, causing cardiac tamponade, and large amount of hemorrhagic fluid within the thoracic cavity, causing atelectasis. Blood was not identified in the abdomen at the time of necropsy; the report of blood in the abdomen prior to death most likely represents inadvertent sampling of pericardial or thoracic fluids through the abdominal wall and diaphragm. This wolf had extensive chronic pericarditis and epicarditis which, when combined with fibrinoid arteritis of the coronary vessels, was the cause of this wolf's pericardial effusion. A definitive underlying etiology for this pericardial and epicardial inflammation was not identified grossly, histologically on H&E or in numerous special stains. Multiple bacterial species were isolated on aerobic and anaerobic culture of a pericardial swab; however, there were no large colonies of bacteria present, no significant accumulations of fibrin and minimal neutrophil infiltration on histologic

examination which makes these isolates more likely to represent postmortem overgrowth or contaminants. The presence of hematoidin in the tissue sections indicates that the blood in the pericardial sac had been present for some time prior to death and did not represent acute hemorrhage into the pericardial sac. The chronic presence of blood in the pericardial sac, causing cardiac tamponade, and the numerous other cardiac findings (left ventricular myocardial infarction, degenerative valve disease, and membranous interventricular septum) likely resulted in decreased cardiac output and accumulation of fluid in the thoracic cavity.

mjs

Dictated by: ANIBAL G. ARMIEN, DVM, MSc, PhD, Diplomate, ACVP, PATHOLOGIST on 5/20/2015 4:19 PM

Attending Specialist: MATT STUROS, DVM, PATHOLOGY RESIDENT; ANIBAL G. ARMIEN, DVM, MSc, PhD, Diplomate, ACVP, PATHOLOGIST

Electronically Signed By: ANIBAL G. ARMIEN, DVM, MSc, PhD, Diplomate, ACVP, PATHOLOGIST on 5/26/2015 11:45 AM

Testing Summary

Laboratory/Procedure	Ordered	Count Result	Quantifier	Interpretation	Result Value Entered	Completed
Bacteriology						
Aerobic Culture-Heart	05/14/2015	1 E. coli Non-haemolyt	3+	Mixed culture	05/18/2015	05/18/2015
Aerobic Culture-Heart	05/14/2015	1 Klebsiella sp.	3+	Mixed culture	05/18/2015	05/18/2015
Anaerobic Culture-Heart	05/14/2015	1 Bacteroides sp.			05/22/2015	05/22/2015
Anaerobic Culture-Heart	05/14/2015	1 C. perfringens	2+		05/18/2015	05/18/2015
Anaerobic Culture-Heart	05/14/2015	1 C. sordellii			05/22/2015	05/22/2015
Anaerobic Culture-Heart	05/14/2015	1 Clostridium sp.		Unable to speciate	05/22/2015	05/22/2015
Anaerobic Culture-Heart	05/14/2015	1 Peptostreptococcus			05/22/2015	05/22/2015
Histology						
Fite's Stain-Tissue	05/20/2015	2 Slide Prep Complete			05/20/2015	05/20/2015
Gram Stain-Tissue	05/18/2015 - 05/20/2015	2 Slide Prep Complete			05/18/2015	05/20/2015
Grocott's Methenamine Silver Stain-Tissue	05/20/2015	2 Slide Prep Complete			05/20/2015	05/20/2015
H&E Slide Preparation-Tissue	05/15/2015	13 Slide Prep Complete			05/15/2015	05/15/2015
Unstained Paraffin Sections-Tissue	05/18/2015 - 05/20/2015	10 Slide Prep Complete			05/18/2015	05/20/2015
Immunohistochemistry						
Calretinin (IHC)-Tissue	05/20/2015	1 Slide prep complete			05/21/2015	05/21/2015
Cytokeratin MNF 116 CKs5,6,8,17,19 (IHC)-Tissue	05/18/2015	1 Slide prep complete			05/18/2015	05/18/2015
CD18-canine (IHC)-Heart	05/20/2015	1 Slide prep complete			05/20/2015	05/20/2015
Lyme Disease (IHC)-Tissue	05/18/2015	1 Slide prep complete			05/18/2015	05/18/2015
Myeloid Histiocyte Antigen IHC-Tissue	05/18/2015	1 Slide prep complete			05/18/2015	05/18/2015
Vimentin (IHC)-Tissue	05/18/2015	1 Slide prep complete			05/18/2015	05/18/2015
Necropsy						
Necropsy/Gen Exam per Animal-Not applicable	05/14/2015	1 N/A			05/14/2015	05/14/2015
Necropsy/Histopathology Only						
Histopathology-Tissue	05/14/2015	1 See report			05/14/2015	05/14/2015