Hi-Ho Liver, Away!

Infectious canine hepatitis

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Introduction

Infectious Canine Hepatitis is a severe disease in canids and ursids caused by Canine Adenovirus 1 (CAV-1). It is most common in animals less than 1 year old and characterized by a subclinical to rapidly fatal disease that causes severe necrosis of the liver, widespread hemorrhage ranging from petechia to fulminant bleeding, corneal edema, and less commonly neurologic signs.[9] This disease is more severe in younger animals and is considered to be a rare disease due to widespread vaccination. In older or vaccinated animals, infection with CAV-1 may be subclinical or only cause a mild pharyngitis. A modified live CAV-1 is available however due to its unique type III hypersensitivity reaction, a modified live Canine Adenovirus 2 (CAV-2) vaccine is almost exclusively used for its cross-protective properties.

History and Presentation

Kemosabe is an approximately 5-month-old, intact, Treeing Walker Coonhound that presented to Mississippi State College of Veterinary Medicine Internal Medicine department on 1/14/2020 after referral from his referring DVM for worsening lethargy, anorexia, and thrombocytopenia. Kemosabe is an indoor/outdoor dog that lives with 3 other dogs (2 adults and one puppy). He presented to his primary veterinarian with a 3-day history of lethargy and anorexia after the owners noted he walked into his kennel to lay down and would not come out or eat, there was no vomiting or diarrhea. The owners reported that he was not up to date on his vaccines and had not been dewormed. They expressed a concern about possible poisoning by their neighbors. On presentation he was depressed, he had pale and tacky mucous membranes with a capillary refill time of greater than 3 seconds. His third eyelids were prolapsed, his hair coat was dirty and a few ticks were noted. He was weak and did not want to stand. No abnormalities were appreciated on cardiac auscultation. He had increased lung sounds with no

crackles or wheezes. On abdominal palpation his abdomen was soft and non-painful with a few gas distended and thickened loops of bowel. Due to his history of being behind schedule on vaccines and not being dewormed, a fecal and parvovirus test (most likely a snap ELISA test for parvovirus antigens) was performed. A 4DX snap test (a snap ELISA that tests for heartworms, ehrlichiosis, Lyme disease, and anaplasmosis) was also performed due to the multiple ticks removed. All tests were negative. A Complete Blood Count (CBC) and blood chemistry panel were performed, radiographs were declined at that time.

The first CBC and blood chemistry panel revealed a mildly decreased hematocrit of 34.7% (37.0-55.0), a severely decreased thrombocyte count at 5,000 (165,000-500,000), and mild lymphocytopenia at 0.68 x 10^9 (1.00-4.80 x 10^9). His chemistry panel showed an increased ALT at 344 (10-90) and an increased phosphorus at 7.1 (2.5-5). All other values were within normal limits. An intravenous catheter was placed and Kemosabe was treated with sodium chloride at 30mls/hr and intravenous lipids at 1.5ml/kg over fifteen minutes (15mls) which was then decreased to 0.25ml/kg/min for an hour (2.5mls/min). The next morning (1/14/2020) he was depressed and did not want to stand, the referring veterinarian noted that he had not moved much overnight but he was eating. A second CBC and blood chemistry were performed and showed a worsening thrombocyte count with 0 thrombocytes being reported which was confirmed with 0 platelets seen on a blood smear. The mildly decreased hematocrit and lymphopenia were static at 34.9% and 0.75 x $10^{9,}$ respectively. The blood chemistry panel revealed that the ALT continued to increase at 769. The ALP was increased at 165 (11-140) and total bilirubin was mildly increased at 1.0 (0.2-0.6). All other values were within normal limits. Abdominal radiographs were performed at this time but there were no abnormal findings seen. The referring veterinarian discussed a poor prognosis with the owners and recommended referral. The owners were

interested in pursuing necropsy if the dog passed so they were referred to MSU-CVM for a possible platelet transfusion or necropsy if euthanasia was elected.

Upon presentation to the MSU-CVM Internal Medicine department on 1/14/2019 Kemosabe was dull and depressed. He had a body condition score of 3/9 and weighed 13.4 kg. He was tachycardic with a heart rate of 180 beats per minute, had a rectal temperature of 102.7°F with blood noted on the thermometer, and a respiratory rate of 24 breaths per minute. Mild petechiations were noted on his ventral abdomen, inguinal area, and prepuce. He had a mild amount of gingival bleeding and his bottom right canine (404) was grey and dead. During the exam he urinated and hematuria was appreciated. At this time, all further diagnostics were declined and Kemosabe was humanely euthanized and submitted for necropsy

Necropsy Findings

Upon external examination this dog has a body condition score of 4/9. There is a 6 cm shaved area on the cranial aspect of the right antebrachium proximal to the carpus (consistent with antemortem intravenous catheter placement). There are few scattered pinpoint red foci around the tip of the prepuce and on his inguinal area (petechiation). His deciduous canines, molars, and premolars are all still present and the gingiva over his upper canines and premolars is reddened.

The gross findings are generally unremarkable with multiple mild lesions noted. The subcutaneous fat is tacky and normal ingesta is present in the stomach and there are multiple intestinal worms (consistent with roundworms, hookworms, and tapeworms). The mesenteric lymph nodes are mildly enlarged. The gallbladder wall is mildly thickened and opaque. The

pericardial sac has approximately 5 mL of red tinged fluid present and both atrioventricular valves are mildly thickened. All other gross findings are within normal limits.

On histopathology the liver is the most affected organ. There are extensive areas where hepatocytes are hypereosinophilic, swollen and have lost cellular detail, consistent with severe, massive hepatocellular necrosis and degeneration. Many of the affected hepatocytes have swollen distorted nuclei with large basophilic intranuclear inclusion bodies (consistent with adenoviral inclusion bodies). Similar inclusions bodies were found within endothelial cells of the renal glomeruli and spleen. Rarely, the brain has congested vessels with a mild lymphocytic perivasculitis and mild rare hemorrhage expanding perivascular spaces. Immunohistochemistry to identify canine adenovirus 1 was submitted to a referred laboratory and had positive staining of the previously described inclusion bodies.

Pathophysiology

Infectious canine hepatitis is caused by Canine Adenovirus I (CAV-1). It is a non-enveloped DNA virus in the genus Mastadenovirus that is spread through ingestion of infectious urine, feces, or saliva.[1] It causes severe liver disease in canids (dogs, coyotes, foxes, and wolves) and bears and is a minor cause of acute respiratory disease. It is now a rare condition due to vaccination; however wild canids are a reservoir for this disease.[10] Initial replication of the virus occurs in the tonsillar crypts and Peyer's patches often causing a mild tonsillitis or pharyngitis. It then spreads to the regional lymph nodes and blood leading to a viremia. This virus has an affinity for endothelium, mesothelium, and hepatic parenchyma leading to a widely disseminated infection.[8] The target organs are the liver and kidneys but often extends to the lungs and spleen. This disease primarily affects younger dogs with an incubation period of 4-9 days and there are three main presentations that may occur following infection. The first is per-acute disease where

the affected animal is often found dead without any apparent illness or after only a short course of symptoms. The second syndrome is acute disease characterized by fever, depression, anorexia, vomiting and bloody diarrhea but may also include petechial hemorrhages of the gums and skin with pale mucous membranes and icterus. More critical cases will have abdominal pain, spontaneous hemorrhage (especially of the oral mucosa), or hematomas. Because of widespread vascular endothelial damage the acute presentation may progress to disseminated intravascular coagulation and is often fatal. The third syndrome is mild disease that usually occurs in vaccinated animals and may only present as mild pharyngitis or tonsillitis.[1]

Infectious canine hepatitis earned its name due to the widespread and extensive destruction of hepatocytes. Bloodwork of animals clinically affected usually shows leukopenia, thrombocytopenia, and prolonged clotting times. Neurological involvement is uncommon but severely affected animals may have convulsions caused by vasculitis in the meningeal vessels leading to hemorrhage, edema, and necrosis or hepatic encephalopathy. While neurological involvement is rare in dogs it is one of the most common signs to occur in foxes earning the original name of the disease, epizootic fox encephalitis. Some animals may also experience temporary paralysis due to the neural damage.[3,7] In approximately 25% of infected animals, bilateral corneal opacity occurs after 7-10 days of infection and usually spontaneously resolves. Following acute infection, the antibody response often results in the formation and precipitation of immune complexes specifically in the cornea and kidney resulting in severe glomerulonephritis and uveitis.[9] In some dogs vaccinated for CAV-1, a type III hypersensitivity reaction occurs resulting in the same corneal edema, endothelial damage, and anterior uveitis commonly referred to as “blue eye” that can be seen following resolution of infection.[10] Older

dogs that contract this virus are more commonly subclinical and may only present with mild pharyngitis or tonsillitis. The overall mortality rate is approximately 20%.

Pathological gross findings often reveal an enlarged liver and spleen, edema (especially of the gallbladder), hemorrhage of superficial lymph nodes and petechial to ecchymotic hemorrhage most often seen on the gingival mucosa. Findings are often inconsistent and may include widespread hemorrhage within the renal cortices, lungs, and rarely the brain and metaphysis of long bones. The liver and spleen may appear mottled due to the widespread necrosis. Typical histopathology shows severe multifocal hepatocellular necrosis within centrilobular regions. Intranuclear viral inclusion bodies can be found within Kupffer's cells, hepatocytes, bile ducts, corneal endothelium, and vascular endothelial cells, especially within the capillaries of renal glomeruli.[5] In Kemosabe's case approximately 50% of the liver was necrotic and intranuclear inclusion bodies were found in nearly every hepatocyte. The histological findings were surprising given that his liver values (ALP, ALT, Total Bilirubin) on his chemistry panels did not reflect that amount of damage present and on gross examination the liver looked normal with only mild enlargement.

Diagnostics

This disease is easily prevented by a good vaccination protocol and due to the vague clinical signs diagnosis is usually made postmortem. The most common diagnostic techniques are viral isolation, PCR, ELISA, or through serology (enzyme immunoassay, hemagglutination-inhibition, or neutralization assay).[2] Immunohistochemistry and immunofluorescence are also available. PCR is the most reliable way to differentiate CAV-1 from CAV-2 and is the most common diagnostic technique.[4] The intranuclear inclusion bodies are diagnostic and the virus

can be isolated in the urine for up to 6-9months following resolution of clinical signs. In subclinical or mild cases tonsillitis is highly suggestive of CAV-1.

Treatment and Prevention

There is no treatment for CAV-1 and supportive care is the only option after an animal has been infected. Supportive care is aimed at maintaining hydration and controlling hemorrhage. These patients very commonly require a plasma transfusion or whole blood transfusion and in Kemosabe's case a plasma transfusion would have been critical. Unfortunately, due to the severe hepatic damage that this virus causes, acute death is common in younger animals. Older animals are typically subclinical and only develop a mild pharyngitis. Corneal edema typically resolves on its own but atropine can help with discomfort. Corticosteroid are contraindicated in infected animals but have been used to treat uveitis caused by the CAV-1 vaccine. Animals that recover can still shed the virus in their urine for up to 6-9 months so they should be quarantined following treatment. Fortunately, this virus is quite rare due to the extremely effective vaccine available.

There is a killed and modified live vaccine available for CAV-1 neither are recommended for different reasons. The modified live CAV-1 vaccine has been shown to cause a type III hypersensitivity reaction in dogs. It results in viral antigen-antibody complexes to precipitate out in the eye, specifically the anterior chamber. It causes anterior uveitis and damage to the corneal endothelium which both result in corneal edema and the classic "blue eye" appearance.[10] In many cases this will resolve by itself but can be treated with atropine or corticosteroids, however; it may be permanent in some cases. Less commonly the type III hypersensitivity reaction will also result in mild nephritis and result in urine shedding of the virus.[8] The killed

vaccine does not cause the same hypersensitivity reaction however it does not have good efficacy.

The alternative is to vaccinate with a modified live CAV-2 vaccine. CAV-1 and CAV-2 are different in morphology and pathogenicity however they are extremely antigenically similar. Due to their antigenic and immunologic similarity, vaccination with CAV-2 is cross-protective for CAV-1 and will result in a sufficient immunity without the blue eye hypersensitivity reaction. The CAV-2 antigen is typically in a combined vaccine with canine parvovirus, canine parainfluenza, and canine distemper virus. The vaccine can be started in dogs as young as 6 weeks and should be repeated at 2-4-week intervals until they are 16 weeks of age. It should be boostered again 1 year after the initial series and lasts for 3 years.[6] This was an important diagnosis for Kemosabe as he lived with another puppy who's susceptibility to this disease depends on their vaccination status. Had Kemosabe been up to date on his vaccines, this disease was unlikely to occur.

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