

DETECTION AND TREATMENT OF A POSSIBLE NEW DISEASE SYNDROME IN A CAPTIVE BLACK RHINOCEROS, (*DICEROS BICORNIS*)

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Introduction

In captivity, the black rhinoceros (*Diceros bicornis*) is a difficult species to manage due in part to many disease problems. In 20 years of maintaining black rhinoceros at the Denver Zoological Gardens we have had 13 births. Presently, our black rhinoceros program consists of one adult breeding pair and a single juvenile male. During this same time period, we have experienced several diseases in our rhinoceros including: hemolytic anemia (Miller, Boever, 1982), leukoencephalomalacia (Miller et al., 1990), salmonellosis (Kenny et al., 1996) and mucosal/skin disease (Munson, 1992). In 1995, our 14 year old male presented with vague symptoms including; straining to urinate, lethargy, increased water intake, and generalized edema which was particularly prominent in the prepuce. This rhino was immobilized several times over the ensuing months to obtain diagnostic information and provide treatment. The exact cause for the animals condition remains unknown although it demonstrated elevated titers for equine herpesvirus (EHV). This report will discuss the diagnostic tests employed, treatment and ongoing management for this animal.

Case report

In November 1995, our 14 year old male showed signs of straining to urinate and depression. Initial etiologies considered either bladder infection or urolithiasis. Treatment was implemented using an antibiotic 960 mg/ 30 tablets trimethoprim and sulfamethoxazole (TMS) (Geneva Pharmaceuticals, Inc. Broomfield, Colorado 80020, USA), an antiparasitic ivermectin (0.2 mg/kg, Eqvalan, Merck & Company Inc., Rahway, N.J. 07065-0912, USA) and a anti-inflammatory 1000 mg flunixin meglumine (Banamine, Schering-Plough Animal Health Corp., Kenilworth, N.J. 07033, USA) which were administered orally in grain, due to the clear discomfort of the animal. These symptoms continued for five days with increasing lethargy. At this time, blood was obtained from the left ear vein without immobilization. Blood chemistry demonstrated, anemia (hematocrit 27% [reference range 36.7 +/- 9.5]), serum biochemistry hypoalbumenemia (1.71 g/dl [reference range 2.1 +/- 0.7]), and hypophosphatemia (2.94 mg/dl [reference range 4.5 +/- 1.2]). These tests were evaluated against what would be considered normal ranges for captive black rhinoceros (Table 1). Treatment consisted of 10 mg/ml iron dextran (Vedco, Inc. St. Joseph, MO 64504) given intramuscularly via pole syringe. On the sixth day after intital presentation the rhino became edematous with swelling occurring ventrally and in the prepuce, and had difficulty breathing and rising. At one point, it had trouble fitting through a 122 cm wide transfer door due to the swelling. The prepuce became so enlarged that walking was

apparently painful for the rhinoceros. Ten ml dexamethasone (Vedco, Inc.) was given intramuscularly to try and reduce swelling. Oral doses of a diuretic furosemide (Lasix, Hoechst-Roussel, Severnville, N.J. 08876-1258) was implemented for added reduction in swelling. TMS was discontinued at this time, because the animal had not improved and there was concern that it might be contributing to its worsening condition.

Two weeks later the rhinoceros showed a reduction in the edema on the ventrum and prepuce. However, the coronary bands of the front feet became very moist, and purulent. The loss of integrity of the tissue and nails was evident. Zoo staff were now concerned the rhino would slough its nails. In addition, the rhino developed multifocal ulcers on the hips and extremities. An immobilization was scheduled at this point for more detailed evaluation including radiographing all feet. Diagnostic tests run were a complete blood count, leptosporosis titers, and serum iron.

The first immobilization occurred three weeks after the initial presentation, then once a week for a period of four weeks. A final immobilization was performed twenty six and a half weeks after initial presentation of symptoms. The rhino was immobilized using M99 etorphine hydrochloride (M99, Wildlife Laboratories Inc., Fort Collins, Colorado 80524, USA) administered via a pole syringe. Additional diagnostic evaluations were performed including ultrasound via the rectum, muscle biopsies and bone marrow samples. The ultrasound visualized the pelvic anatomy and bladder only, but it was interpreted as normal; results from the muscle biopsy showed evidence for a prior vasculitis, and bone marrow aspirate was non-diagnostic. On re-examination, the rhino developed necrotic laminitis in the front feet. This required the removal of the nails and necrotic lamina from the medial toes of both front feet. Portions of the soles of both front feet were also necrotic and had to be debrided. Pressure lesions increased on the bony prominences of the forelegs and chin.

Management

Due to the severity of the rhino's condition extensive management practices were implemented. Intensive hydrotherapy was instituted for at least 20 minutes two times a day using a mixture of Nolvasan and Epsom salts. Cleaning was performed with sterile handbrushes and the solution flushed through nails and undermined areas. It was crucial that the animal's feet remain as clean and dry as possible. In order to achieve proper cleansing and application the procedure was performed in very close proximity to the rhinoceros. Daily application of tincture of iodine sprayed on exposed lamina accelerated hardening of nails. A mixture of 10% neutral buffered formalin (Richard-Allen Medical, 8850 M89, Box 351, Richard, MI 49083-0351, USA), phenol (Gateway Products, Holly , Colorado 81047-0529, USA), and tincture of iodine (Vedco, Inc., Omaha, NE 68127) combined at a ratio of 1: 1: 1 was applied to soles of feet when the animal rested to increase the hardening of pads. After the sores were cleaned, Vitamin A and D ointment (Schering Corporation, Kenilworth, N.J. 07033, USA) was applied to affected areas and immensely helped the healing process. Fortunately, the rhino allowed these procedures to be performed without increased risk to himself or keeper, and in the end was a determining factor in the success of treatment and therapy.

Antibiotics (TMS / 14 tablets b.i.d.) were reinstated seven days after initially being discontinued, along with 23 tablets b.i.d. metronidazole (Flaggal, Geneva Pharmaceuticals, Inc. Broomfield, CO. 80020, USA). They were administered orally mixed in a gruel consisting of one cup bone meal, 12 g Brewers Yeast, mashed bananas, 2 tablespoons honey mixed b.i.d. daily into the grain. Grain and alfalfa was offered when the rhino was undergoing treatments keeping the it calm and stable.

Another consideration was the possible transmission of this unknown ailment to the rest of the rhinoceros collection, thus a quarantine was instituted working within the parameters of the exhibit. Since there are no quarantine facilities in the rhino section, all other rhinos were kept physically removed from the ill animal. Other aspects of management included daily bedding of the stalls with wood chips and hay to decrease the spread of feces and to insure a dry environment.

Due to the rhino's illness, access to the outside yard was not permitted for a period of six months. Behavioral enrichment was implemented for mental and physical stimulation. Several "toys" were developed that could be hung on chains from a secure mount in the ceiling of stalls. Some of the items offered were "boomer balls", tires, logs and a large inverted rubber tub. Implementation of enrichment reduced stereotypical behavior of banging on bars, and increased physical and mental stimulation through interaction with these devices.

Discussion

Prior to the rhino's initial presentation, our collection of Grevy's zebra (*Equus grevyi*), experienced an outbreak of equine herpes in which a young foal succumbed. Equine herpes (EHV-1), or equine herpes rhinopneumonitis has three clinical syndromes associated with this disease: respiratory, abortigenic, and neurological (Liu 1980). The natural reservoir of (EHV-1) is found in the horse. Transmission of this disease occurs through either direct or indirect contact with infected mucosal discharge, aborted fetuses, and placenta or placental fluids (Merck 1991). The respiratory form has an incubation period of 2-10 days followed by these clinical symptoms: fever of 102 -107 degrees Fahrenheit lasting 7-10 days, discharge of mucosa from nose and eyes, coughing, and depression (Blood et al., 1983, Merck 1991, Koterba 1990, Smith 1990, Lui 1990). A thorough examination of zebra and rhinoceros records was conducted to ascertain the time frame of both occurrences. The records showed that the episode of herpes in the zebra herd was exactly three weeks before the rhino presented with symptoms. At this point, freezer banked serum from 1988, was sent for analysis to determine if equine herpes had been present historically in the rhino. Test results were negative. Another immobilization was scheduled, an evaluation of the progress of the feet was conducted, as well as, more freezer banked serum samples from the rhino were evaluated for equine herpes. Other samples were sent to National Veterinary Services Laboratories Ames, Iowa (NVSL) to evaluate the following equine viral diseases: Bluetongue enzyme-linked immunosorbent assay (Elisa), Epizootic Hemorrhagic Disease (AGID), Equine Infectious Anemia (AGID), Equine Viral Arteritis (SN 1:4), and Malignant Catarrhal Fever (IP & SN). The results from these tests proved negative, but the blood chemistry showed a suggestion of elevated titers for equine herpes.

Several factors were investigated for the possible transmission of equine herpes to the rhinoceros. A communal refrigerator was located on the rhino side of the Pachyderm building where diets for hoofed stock including the Grevy's zebra were stored. Diets would be retrieved by keepers through out the day. Transmission could have occurred by tracking feces on the soles of boots. Another factor considered was that the relief keeper for rhinos also worked with the Grevy's zebra collection, and may have transferred fecal matter via coveralls or boots. Once diagnostic tests proved positive titers for equine herpes, steps were taken to reduce traffic from the Pachyderm building to other areas with hoofed stock. Footbaths were set up on the rhino side of the building entering and leaving the section. The refrigerator was removed and set up within the main hoofed stock area to eliminate the necessity of

transferring between work areas. In addition, the keepers had separate coveralls and boots for working with the rhinos which were left in the section when finished. Recently, information has been received that two other institutions housing black rhinoceros, each have had a rhino presenting with similar symptomology. Common factors for each case have been localized edema, anemia and vasculitis with hemorrhage between muscle fibers found on muscle biopsies. These animals have been evaluated for equine herpes, and did not show elevated titers for this disease.

Conclusion

After six separate immobilizations on the rhinoceros, the cause for the animals condition remains unknown, but suggestive of a case of equine herpes. At this point, the rhino has made a full recovery with new nail growth occurring on affected front feet, lesions have completely healed, and the hematocrit is within normal ranges (46%). Six months later, access to the outside yard was approved after the last immobilization. The question remains, was there a transmission of equine herpes to a rhinoceros, or are we looking at a previously unknown form of rhino herpes.

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Acknowledgments

Special thanks and gratitude are extended to the following people for their guidance and support in the writing of this paper. Dr. Dave Kenny, Dr. Beth Bicknese, Dr. Jeff Baier, Joan Poston, John Wortman and Megan Phillips.

	Hematocrit	Albumin	Phosphorus
Reference	36.7 +/- 9.5	2.1 +/- 0.7	4.5 +/- 1.2
TIME			
5 days after	27%	1.71 g/dl	2.94 mg/dl
Initial			
symptoms			
Two weeks	20 %	2.20 g/dl	4.77mg/dl
Three weeks	18.5%	2.61g/dl	7.03 mg/dl
Four weeks	20%	2.25g/dl	4.44mg/dl
Five weeks	23%	2.2g/dl	4.1mg/dl
Seven weeks	35%	2.40g/dl	4.3mg/dl
Twenty six	46%	2.40 g/dl	4.4 mg/dl
weeks			

Table 1. Hematology and selected chemistry values from initial presentation of illness of adult male black rhinoceros through twenty six weeks in comparison with normal values found in Average Physiological Values, (International Species Information System, Apple Valley, MN 55124-8151, USA) June 1992.

6/4/88	< 1:2
12/12/95	1:256
16/1/96	1:128

Table 2. Freezer banked serum for evaluation of equine herpes was sent to (CSU) Colorado Veterinary Diagnostic Laboratories, Colorado State University, Fort Collins, CO 80523-1644, USA. Methodology used to evaluate equine herpes was serum neutralization.