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CORONAVIRAL ENTERITIS IN AN OSTRICH (*STRUTHIO CAMELUS*) CHICK

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Abstract: Coronaviral particles were identified in intestinal contents and epithelial cells of the lower small intestine of a dead 18-day-old ostrich (*Struthio camelus*) chick. This ostrich chick was one of two with a 1-wk history of anorexia, lethargy, weakness, and diarrhea. Histologic changes included villous atrophy, necrotic cells in crypts, and occasional crypt collapse in the lower small intestine. Other lesions were dilation of the proventriculus, nutritional osteodystrophy of the proximal tibiotarsal bones, and degenerative myopathy of the pectoral muscles.

Key words: Coronavirus, enteritis, ostrich, *Struthio camelus.*

INTRODUCTION

Reports of coronaviral enteritis in birds have been limited to turkey poults, where clinical signs of this disease (bluecomb) include inappetence, weight loss, and diarrhea. Outbreaks of coronaviral enteritis have occurred in all ages of turkeys; mortality is higher in poults <6 wk of age.[13] Experimental turkey coronaviral infection is characterized by depressed rate of weight gain, reduced feed consumption, and diarrhea.[9] The incubation period averages 2–3 days. This paper describes a case of coronaviral enteritis in an ostrich (*Struthio camelus*) chick.

CASE REPORT

Two 10-day-old ostrich chicks were presented to the College of Veterinary Medicine at Kansas State University with a history of anorexia, lethargy, and weakness since hatching. Weights of the two chicks at hatching were 1,136 g (chick 1) and 909 g (chick 2). The chicks ate very little for 3–6 days posthatching; when they did, they would eat only boiled egg whites. Two chicks with similar clinical signs had died previously at 8 and 12 days posthatching, and two other chicks died 3 days posthatching.

On physical examination, the chicks were approximately 5% dehydrated and weighed 799 g (chick 1) and 884 g (chick 2). Both chicks had loose droppings. A complete blood count and serum chemistry profile were performed on both chicks prior to initiation of treatment and were compared with normal values.[2,10] Both chicks had hypercreatinemia (0.4 mg/dl), hypoalbuminemia (2.0 and 1.0 g/dl), low albumin : globulin ratios (0.54 and 0.32), and elevated aspartate aminotransferase (303 and 323 IU/L). Chick 1 had hyperproteinemia (5.7 g/dl); chick 2 had hyperglycemia (352 g/dl), hyperkalemia (6.0 mmol/L), and elevated uric acid (16.8 mg/dl). Total leukocyte counts were within normal limits (7,790 and 4,495/mm^3) and included for chicks 1 and 2, respectively, 48 and 65% heterophils (with many degranulated cells), 30 and 11% lymphocytes, 16 and 22% monocytes, and 1 and 2% basophils. Eosinophils were 1% for chick 1 and not counted for chick 2 because of marked degranulation of granulocytes. Packed cell volumes were 34 and 42%. Nonhemolytic *Escherichia coli* was isolated in pure culture from cloacal swabs of both chicks. No parasite ova or protozoa were detected in the feces of either chick.

During hospitalization (2 days), both chicks were tube fed six times daily with 30 ml of a mixture of canned canine maintenance diet, vitamins, and water as a gruel. They also received 40 ml lactated Ringer's solution s.c. b.i.d., 11 mg/kg trimethoprim and 55 mg/kg sulfamethoxazole (Septra Suspension, Burroughs Wellcome Co., Research Triangle Park, North Carolina 27709,

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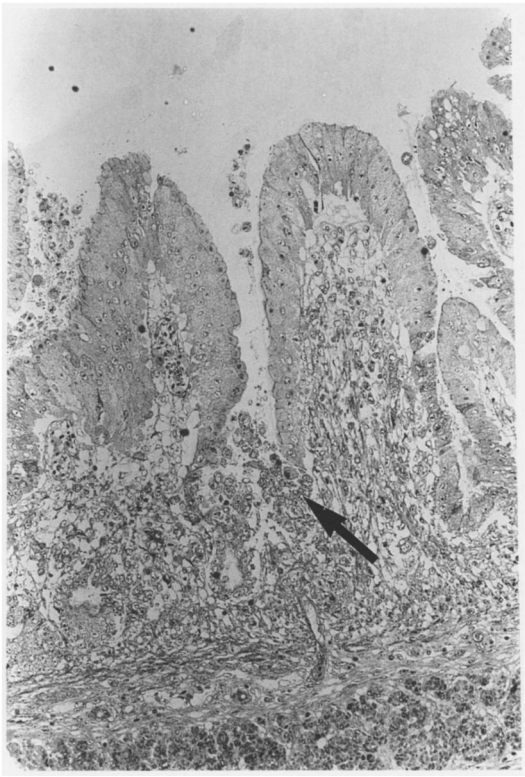

Figure 1. Lower small intestine of an ostrich chick with coronaviral enteritis. Note villous atrophy, eosinophilic inclusions in the apical cytoplasm of villous epithelial cells, and necrotic cells (arrow) in crypts. H&E, ×55.

USA) p.o. b.i.d., 1 ml bismuth subsalicylate (Pepto-bismol, Procter and Gamble Co., Cincinnati, Ohio 45202, USA) p.o. b.i.d., injectable B complex, and supplemental heat (29–31°C). All treatments were to be continued at home for an additional week.

Chick 2 died 7 days after discharge from the hospital and was necropsied. Tissue samples were fixed in 10% neutral buffered formalin and processed routinely for histologic examination (H&E). The mucosa of formalin-fixed lower small intestine was later trimmed to 0.5-cm^2 sections, rinsed several times with 0.1 M sodium cacodylate buffer (pH 7.4), immersed in 2.5% glutaraldehyde and 2% paraformaldehyde in 0.1 M sodium cacodylate buffer (pH 7.4), and processed routinely for transmission electron microscopy. Intestine and liver were collected for bacteriologic examination, and intestinal contents were examined by negative contrast electron microscopy.

Grossly, the proventriculus was dilated to approximately 6 cm in diameter, thin walled, and filled with ingesta. The gizzard was empty. The lower small intestine had a mildly thickened wall and pasty tan contents. The kidneys were mottled pink and white. Both tibiotarsal bones were soft, and a 0.5–1.0-cm-wide band of cartilage extended from the growth plate of the proximal tibiotarsus into the metaphysis.[5]

Histologically, the distal small intestine often had atrophied villi, crypts containing necrotic cells, and occasional crypt collapse (Fig. 1). Eosinophilic inclusions were detected in the apical cytoplasm of many enterocytes. Proventricular mucosa was thinned and had few glands. Hepatocytes were moderately swollen and had increased intracytoplasmic clear spaces. Pectoral myocytes had moderate multifocal degeneration, necrosis, and mild mineralization (nutritional myopathy). The proximal tibiotarsus had a moderately thickened periosteum, and the cortex was thin and only partially mineralized. Long projections of mineralized cartilage extended into the metaphysis with no detectable osteoid or active osteoblasts. The bursa of Fabricius had few lymphoid cells. No significant lesions were present in brain, heart, kidney, thyroid gland, parathyroid gland, pancreas, adrenal gland, ventriculus, or trachea.

Ultrastructural examination of negatively stained intestinal contents contained moderate numbers of virus particles varying in diameter from 50 to 150 nm (Fig. 2). The particles had distinctive closely packed surface projections typical of coronaviruses. Similar virus particles were detected by transmission electron microscopy within vesicles of, and adjacent to, crypt epithelium (Fig. 3). Virus-infected cells occurred in small clusters and were often widely separated. The apical cytoplasm of many enterocytes contained aggregates of fibrillar material, which corresponded with the

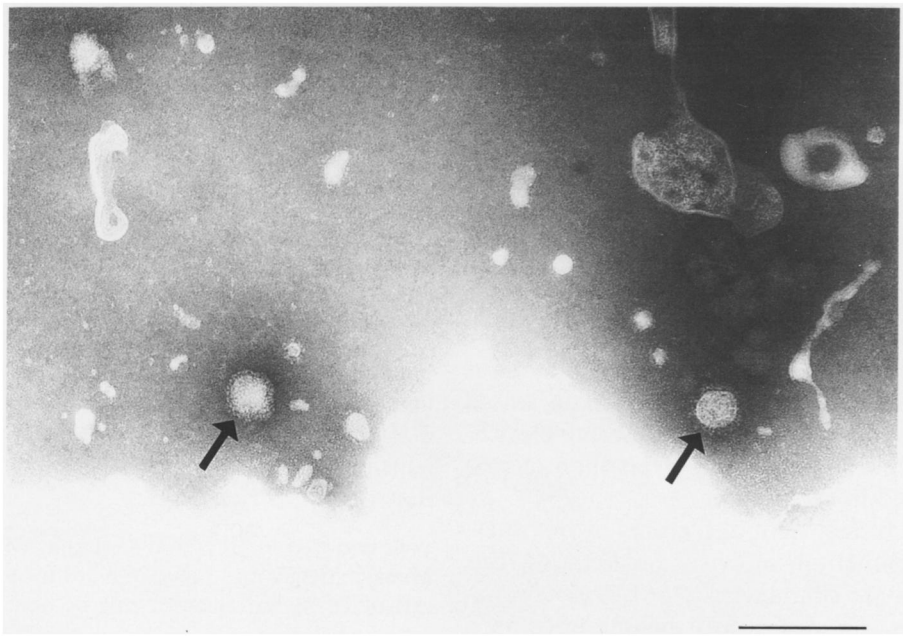

Figure 2. Negatively stained electron micrograph of lower small intestinal contents of an ostrich chick with coronaviral enteritis. Note coronaviral particles (arrows) with surface projections. Bar = 0.2 μm.

intracytoplasmic inclusions seen on light microscopy.

Abundant nonhemolytic *E. coli* was isolated from intestine and liver. Abundant *Aeromonas* sp. was isolated from intestine and group D *Streptococcus* sp. from the liver.

DISCUSSION

Light microscopic changes in the intestine of this ostrich chick resembled those of coronaviral enteritis in other animal species. Typical lesions shared by various mammalian species include shortened intestinal villi, villous fusion, and flattening and necrosis of villous epithelium, especially in the distal small intestine.[8] Calves with coronaviral enteritis often have dilated colonic crypts of Lieberkühn containing intraluminal necrotic cells, similar to the changes noted in the ileum of this ostrich chick.[11,12] In contrast, villous atrophy and villous epithelial desquamation, but no crypt lesions, were described in experimentally infected turkey poults.[6]

Coronavirus infects villous epithelium of the small intestine and both crypt and surface epithelium of the colon of experimentally infected calves.[11,12] Similarly, both villous and crypt epithelium of turkey embryos are infected.[14] As a result of this epithelial

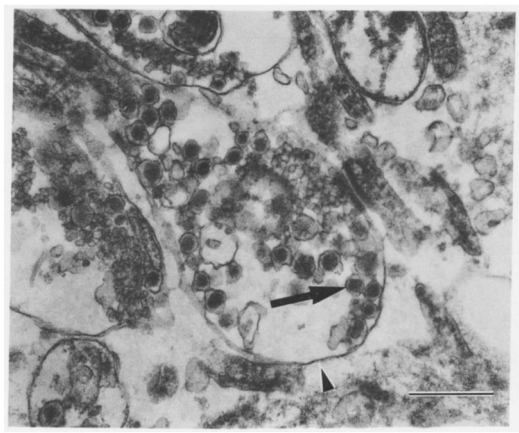

Figure 3. Transmission electron micrograph of epithelial cell from an ostrich chick with coronaviral enteritis. Note cluster of coronavirus particles (solid arrow) within a vesicle (arrowhead). Bar = 0.5 μm.

infection, the reduced absorptive and digestive capacity of the intestine (because of the immature villous epithelium) and the impaired lumen-to-extracellular space fluid flux of sodium are thought to result in the diarrhea associated with the disease.[11,16] In pigs, the poorly digested and unabsorbed ingesta undergo bacterial degradation and fermentation in the intestine; osmolality of the intestinal contents is consequently increased, and fluid is drawn into the intestine by the resulting osmotic gradient.[7] Villous epithelial changes and associated decreased absorption likely explain the serum electrolyte abnormalities and hypoalbuminemia detected in the present case.

The ultrastructural features of the intestinal epithelium and virus isolated from this ostrich are characteristic of turkey coronaviruses. These features include enveloped virus particles budding into membrane-lined cisternae in the cytoplasm of epithelial cells and abnormal amounts of fibrillar material in some cells.[1]

In experimentally infected poults, coronavirus particles were identified ultrastructurally in the contents of ceca and bursa of Fabricius.[15] The spleen of coronavirus-infected turkey poults is often smaller than normal.[13] Lymphoid cells in the bursa of Fabricius in this ostrich chick were severely depleted, possibly as a result of the coronavirus infection.

Leg deformities are very common in ratites and may be associated with muscle necrosis (nutritional myopathy) or nutritional osteodystrophy (rickets) and osteomalacia[2] and with high-protein diet, rapid growth, and lack of exercise. Anorexia and malabsorption in the present case likely contributed to leg lesions. The dilated proventriculus may have resulted from frequent tube feeding.

Coronavirus can be an important cause of disease in avian species. Coronavirus was identified in a recent study[4] as the most common enteric virus associated with outbreaks of diarrhea in turkey poults. No cross-reaction was detected between turkey coronaviruses and antisera against coronaviruses of avian infectious bronchitis, porcine transmissible gastroenteritis, bovine neonatal calf diarrhea, or mouse hepatitis.[3] No experiments were performed to study cross-reactivity of the ostrich coronavirus with other coronaviruses. Turkey coronavirus was not transmissible to chickens, pheasants (*Phasianus colchicus*), gulls, coturnix (*Coturnix coturnix*), or quail (*Colinus virginianus*).[14] The ostrich coronavirus may be a new species of coronavirus. This case is the first report of a coronavirus in the ostrich. Coronaviral enteritis should be considered in the differential diagnosis of diarrhea in ostrich chicks.

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