**DISSEMINATED VISCERAL COCCIDIOSIS IN A WHITE-NAPE CRANE**

*(Grus vipio)*

**GERRY M. DORRESTEIN**\(^1\) and **JUDITH M.A. VAN DEN BRAND**\(^2\)

**Affiliation:**
1. Diagnostic Laboratory of the Dutch Research Institute for Avian and Exotic Animals (NOIVBD), Wintelresedijk 51, 5507 PP Veldhoven, The Netherlands. [Dorresteingm@NOIVBD.nl](mailto:Dorresteingm@NOIVBD.nl)
2. Department of Pathobiology, Division Pathology, Faculty of Veterinary Medicine. Utrecht University, Yalelaan 1, 3584 CL Utrecht, The Netherlands

**Abstract**
This presentation reports of a case of disseminated visceral coccidiosis (DVC) in a 4-month old captive white-naped crane (*Grus vipio*) in The Netherlands. Based on this case a review is presented describing macroscopic and histopathologic changes, therapeutic possibilities. It also presents experimental derived information about the cycle of *Eimeria gruis* and *E. reichenowi*, as well as molecular biological differences that demonstrated these parasites form their own cluster, which classifies them separately from other *Eimeria* spp.

**Introduction**
In cranes, coccidia that normally inhabit the intestine sometimes become widely distributed throughout the body (Carpenter et al, 1980; Parker and Duszynski, 1986; Friend and Franson, 1999). The common coccidial parasites of Gruidae are *Eimeria gruis* and *E. reichenowi* (Courtney et al, 1975; Forrester et al, 1978; Augustine et al, 1998). The resulting disease, disseminated visceral coccidiosis (DVC) of cranes, is characterized by nodules consistent with granulomas, on the surface of organs and tissues that contain various developmental stages of the parasite. Lesions of DVC were first seen in captive sandhill cranes (*Grus canadensis*) in the US in the late 1970s. Since then, mortality of captive sandhill and whooping cranes (*Grus americana*) has been attributed to DVC. The disease has also been found on migratory sandhill cranes at several locations, and it is a recurring problem in the only free-ranging population of the nonmigratory Mississippi sandhill crane that reside at the Mississippi Sandhill Crane National Wildlife Refuge in Mississippi (Friend and Franson, 1999). Recently the disease is also documented in a 4-month old Japanese white-naped crane (*Grus vipio*) in a zoo in Korea (Kim et al, 2005). There is no documented report on DVC in the European situation.

**Case Report**
A 4-month old female white-naped crane (*Grus vipio*) was parent-incubated and hatched as a single chicken in the Dutch Foundation for Refuge and Care of Parrots (Stichting NOP) in Veldhoven, The Netherlands. It died without noticed clinical symptoms. At necropsy, the most remarkable changes found were a severe hyperaemia and oedema of both lungs that contained almost no air and sunk in water. The liver was swollen with small red and pale well-demarcated foci and the spleen dark red and slightly swollen. There was no fat on the heart or any other place in the body and the right ventricle was slightly dilated. In the small intestines several large nematodes (5-6 cm) were found. These nematodes were determined...
as *Toxacara canis*.\(^1\) The last part of the intestines (10 cm till 2 cm before the cloaca) showed a congested mucosa.

At cytology the impressions of the liver, spleen and lungs showed many schizonts as well as many different stages of coccidial development, intra- as well as extracellular (Figure 1, E and F). There were a high percentage of juvenile erythrocytes. In the intestines many different rod-shaped bacteria were present. These bacteria were considered to be the normal intestinal flora and were not cultured. From the liver a sporadic colony of *Aeromonas* was cultured.

At histology many coccidial stages were present in the kupffer cells and other RES-cells in the sinusoids and blood vessels of the liver and in macrophages in the spleen and lungs. In other organs incidental coccidial stages were found (Figure 1, A and C). In the intestines the coccidial stages were present in blood capillaries in the lamina propria, in macrophages in the blood vessels in the intestinal wall and final coccidial stages in the mucosal epithelial cells (Figure 1, B and D). Especially in the lungs the presence of the coccidial stages resulted in severe oedema and a mixed inflammatory response. The spleen and the bursa were totally depleted of lymphoid cells.

As a coincidental finding nematodes (*Capillaria* sp) were present under the koilin layer of the gizzard. The adult female worms produced eggs with two pole caps that were shed through the koilin layer

The final diagnosis was disseminated visceral coccidiosis.

**Discussion**

Although this is the first published report of disseminated visceral coccidiosis in captive cranes in Europe, it is not the first case seen by the author (GMD, personal data). In the past several cases have been diagnosed in different zoos in The Netherlands. In our case the macroscopic lesions were consistent with descriptions of DVC in the cited literature. The death is associated with an overwhelming systemic infection by the intracellular protozoan parasite, which resulted in enteritis, granulomatous bronchopneumonia, hepatitis, splenitis, and myocarditis (Carpenter et al, 1980; Kim et al, 2005). The macroscopic lesions of DVC in cranes are typically described as small (usually less than 5 millimetres in diameter), raised, light-coloured granulomas. These nodules may be found on any surface within the body cavity, but they are commonly seen on the lining of the oesophagus near the thoracic inlet area and on the inner surface of the sternum. Light-coloured patches may also appear on the surface and within organs such as the heart and liver (Friend and Franson, 1999; Novilla and Carpenter, 2004). As in our case, coccidian parasite-laden macrophages are commonly found in the blood vessels of these organs. These findings suggest that an initial *Eimeria* sp. intestinal infection spread to other organs through the blood vessels. In some cases an immunosuppressed state, suggested by the presence of a *Cryptosporidium* sp infection, possibly contributed to a rapid haematogenous distribution (Kim et al, 2005). In our case the immunosuppression was suggestive by the absence of lymphoid cells in the spleen and bursa.

This case report shows that the respiratory changes were most probably the reason for dying. In an experimental infection in sandhill and whooping cranes no clinical signs attributed to a respiratory infection. Necropsy of naturally infected adult birds revealed granulomas in many organs, including the lung, air sacs, trachea and nares. Experimentally infected sandhill cranes and whooping crane chicks that died from DVC had congestion and consolidated areas in the lung with frothy fluid in the airways. Grossly visible nodules were observed from 10 days post inoculation (p.i.). Granulomatous pneumonia and tracheitis were observed with light microscopy. Lesions were associated with merogonic and gametogonic stages of eimerian coccidia. Oocysts and gametocytes were found in the intestines by 12 days p.i. and in the oesophagus, trachea, bronchi, and lung by 14 days p.i. indicating that crane eimerians can complete their life cycle at these sites. Of the few eimeriid coccidia that

---

\(^1\) In 2007 these worms were redetermined as *Porrocaecum ardeae*, the classical roundworm of cranes (GMD).
have extraintestinal stages of development in birds and mammals, only the species in cranes complete their life cycle in both the digestive and respiratory tracts (Novilla et al, 1989; Novilla and Carpenter, 2004).

To define the area of the intestine that was invaded by sporozoites of *E. gruis* and to study the early development of *E. gruis* in the intestines and visceral organs of cranes monoclonal antibodies, elicited against *Eimeria* spp. of chickens and turkeys, were used on tissues of Florida sandhill cranes. At 6 hr p.i., *E. gruis* sporozoites were found primarily in the jejunum from just proximal to Meckle's diverticulum to the ileocecal juncture. Fewer sporozoites were found in the caeca and rectum, and none were found in the duodenum. Most of the sporozoites were in the middle third of the villi and within the lamina propria. At 14 days p.i., developmental stages were detected in the caeca, jejunum, liver, and lungs but not in the heart, kidney, or brain (Augustine et al, 1998).

Mortality in natural infection has been seen already in 13- to 18-day-old whooping cranes, but also a 9-year-old whooping crane died in outdoor pens at the Patuxent Wildlife Research Centre (Carpenter et al, 1980).

Disseminated visceral coccidiosis is not only a problem in captive cranes. Similar lesions were observed in wild sandhill cranes throughout parts of Midwestern United States, Alaska, and Saskatchewan. These studies revealed a wide geographic distribution and a high frequency of occurrence of DVC in wild cranes (Carpenter et al, 1984).

*Eimeria gruis* and *E. reichenowi* develop at multiple organs or tissues in infected cranes, thus, in spite of morphologic similarity, lacking the specificity of infection sites shown by other *Eimeria* spp. Matsubayashi et al (2005) isolated oocysts of *E. gruis* and *E. reichenowi* from crane faeces in a wintering area in Japan. By phylogenetic analysis, they first demonstrated that partial sequences of the isolates formed their own cluster, which is classified separately from other *Eimeria* spp.

Oocysts can rapidly build up in the environment when birds are overcrowded and use an area for a prolonged period of time. The disease risk increases significantly when these conditions result in oocyst contamination of food and drinking water. In captive situations, good husbandry and sanitation, including continual removal of contaminated feed and litter, can minimize the potential for coccidiosis. Captive birds can be treated with therapeutic agents that control, but that do not eliminate, the level of infection. In an experimental study to test the anticoccidial efficacy of amprolium (at 2.2 ppm), clazuril (at 1.1 and 5.5 ppm), and monensin (at 99 ppm) in sandhill cranes infected with a mixture of *Eimeria* spp. oocysts, monensin was the only anticoccidial drug that provided protection against experimentally induced disseminated visceral coccidiosis (Carpenter et al, 2005).

Light infections result in a substantial level of immunity to that species of coccidia and are use-full in preventing epizootics from this disease. Therefore, the objective is not to completely eliminate infection with coccidia; instead, the focus should be on preventing heavy infections and the establishment and persistence of high levels of environmental contamination with coccidia. For free-ranging birds, flock dispersal may be warranted when overcrowding continues for prolonged periods of time (Friend and Franson, 1999).

---

2 The presence of the *Toxocara canis* most probably is explained by a defect in the sewer system in that exhibit. Dogs are allowed in this bird park and faeces of dogs are collected and dropped in the toilets. The chick most probably selected the worms from dog faeces by eating it.

In 2007 these worms were redetermined as *Porrocaecum ardeae*, the classical roundworm of cranes (GMD)
References


Figure 1. Disseminated visceral coccidiosis in a white-naped crane (Grus vipio), histopathological (A-D) and cytological (E and F) findings.

A. Lung HE obj. 40x, B. Small intestine mucosa HE obj. 40x, C. Liver HE obj. 40x, D. blood vessel in intestinal wall HE obj. 40x (bar = 50 μm), E. Liver Hemacolor obj. 100x, F. spleen Hemacolor obj. 100x (bar = 10 μm).