ATYPICAL NON-ALIMENTARY CRYPTOSPORIDIOSIS IN THREE LIZARDS

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Abstract: Atypical, non-alimentary cryptosporidiosis was diagnosed in two green iguanas, Iguana iguana, and in a Parson's chameleon, Calumma parsoni. Renal cryptosporidial infection was identified in one of the iguanas and in the chameleon. Cryptosporidial sialoadentitis was identified in the other iguana.

Key words: green iguana, Iguana iguana, Parson's chameleon, Calumma parsoni, Cryptosporidium, renal cryptosporidiosis, kidney, salivary gland
INTRODUCTION


In none of the following three unusual cases of cryptosporidiosis in lizards described here was the gastrointestinal tract involved. Infections in two cases were confined to renal tissues in: one green iguana, Iguana iguana (FLF), and in the Parson's chameleon, Calumma (Chamaeleo) parsoni cristifer (FLF). The diagnosis of the third case was associated with sialoadenitis in a green iguana. (RHG).

Case 1

Formalin-fixed tissues of an adult female common green iguana, Iguana iguana, were sent from HM to FLF for histopathological diagnosis. These tissues consisted of skin, skeletal muscle, cartilage, brain, stomach, small and large intestines, liver and gallbladder, heart, lungs, kidneys and adrenals, ovaries and oviducts, pancreas, spleen, and partially regenerated tail. The iguana had a brief history of inappetance, declining health, weight loss and, immediately prior to presentation, tonoclonic seizures. A whole-blood sample was obtained immediately prior to the iguana's death. The only abnormal clinical chemistry values were hypocalcemia (4.71 mg/dL) and severe azotemia, which was determined by a dipstick method (Azoastix® Ames, IA).

Examination of hematoxylin- and eosin-stained, formalin-fixed tissues processed by routine histological methods disclosed the following pathological abnormalities.

Kidneys: There were extensive foci of typical “star-burst” shaped gouty tophi involving the renal glomeruli, proximal and distal convoluted tubules, and adjacent peritubular interstitium. Most of the older gouty glomeruli were characterized by grossly thickened capsular membranes and distorted glomerular capillary tufts, many of which were replaced by radiating stellate ghosts where microcrystalline urates had occupied space before being lost during histological processing. The tubular lesions (consisting of tubular nephrosis and necrosis) were surrounded by fibrocollagenous connective tissue that separated the tubular lesions into “islands” of affected renal medullary tissue. The lumens of the distal convoluted tubules contained myriad numbers of organisms measuring 2.5 - 5.0 μm in diameter; these were attached to the luminal epithelium.

Liver: Generalized hepatocellular vacuolar lipidosis. Most hepatocytes contained — and were distorted by — one or more clear vacuoles that had contained lipid prior to histological processing (during which non-polar solvents removed the intracytoplasmic lipid droplets). The medial tunics of muscular arteries and arterioles exhibited extensive soft-tissue mineralization.
Lungs

The medial tunics of pulmonary arteries and arterioles were affected with the identical soft-tissue mineralization observed in the hepatic vasculature.

Intestine: Several ascaridoid nematodes, hookworms, and pinworms were identified within the jejunum and sacculated colon. Mild nonsuppurative jejunitis was present and may have been associated with the helminthiasis.

The balance of the tissues were unremarkable and did not contribute to the diagnosis of this iguana's terminal disease.

Case 2

An eight-year-old male Parson's chameleon, *Calumma (Chamaeleo) parsoni cristifer*, had been in captivity for at least five years and was housed with an adult conspecific female in a screened outdoor breeding cage. Both chameleons were fed a diet of varied commercially cultured insects every other day. On the day of its death, the chameleon was mildly dehydrated, had regurgitated once, and had passed hemorrhagic stools; immediately prior to death, its mucosae were pale. At necropsy, the colon was found to filled with clotted blood. THB sent tissues consisting of lung, stomach, small and large intestines, liver, gallbladder, spleen, pancreas, kidneys and adrenals, testes, and coelomic fat bodies to FLF for histopathological diagnosis.

Histopathological examination of hematoxylin- and eosin-stained, formalin-fixed tissues processed by routine histological methods disclosed the following pathological abnormalities.

Kidneys: Myriad numbers of round- to pear-shaped organisms measuring 2.0 - 5.0 μm were attached to the luminal epithelium of the proximal and distal convoluted renal tubules. The glomeruli and renal interstitium were not affected.

Pancreas: Many of the pancreatic exocrine cells contained single or multiple clear vacuoles of lipid. The islets of Langerhans were not affected.

Intestine: The mucosae of the small and large intestines were hemorrhagic, swollen to distortion and, in some areas, were discontinuous because of focal denudation and lifting away from the subjacent lamina propria.

Interestingly, an ELISA test that is specific for *Cryptosporidium* (ProSpecT® Cryptosporidium EIA Assay, Alexon -Trend, Inc., Ramsey, MN) was performed on highly diluted, formalin-preserved urates from this chameleon's cagemate; this test reacted weakly positive. At the date of this writing, the cagemate has remained healthy.
Case 3

A six-year-old female green iguana, *Iguana iguana*, was presented to a veterinarian with an owner’s complaint that a mass at the left mandibular commissure had been present for approximately three months. Six punch biopsies of the mass were obtained and submitted to RHG for histopathology. Subsequently, this mass became locally invasive and appeared to extend into and around the left ear. Two multilobulated soft-tissue masses that measured 1 x 1 x 2.0 cm and 1 x 1.5 x 2.5 cm originating from the corner of the left mandible were surgically excised and submitted to RHG for histopathology.

Microscopic examination revealed salivary glandular tissue composed of branching tubular structures lined by tall columnar epithelial cells that displayed marked hyperplasia; the cell layers were as many as ten cells thick. Prominent lymphoplasmacytic and heterophilic granulocytic infiltrates were identified within the epithelium and submucosa. Large numbers of small round structures measuring 2-3 μm in diameter were present along the luminal surfaces of the epithelial cells as well as free within the tubular lumens. Large numbers of identical structures were observed within the intraluminal mucus. In some areas, the lining epithelium had become eroded and was replaced with histiocytes and multinucleated giant cells.

**DISCUSSION**

Although reptilian cryptosporidiosis usually involves infection of gastrointestinal organs, particularly the stomach (in snakes and some lizards) and intestine (in some lizards), extra-alimentary infection has been reported in an iguana that was affected with an aural polyp subjacent to the tympanic membrane (Fitzgerald, *et al.*, 1998). The speciation of the *Cryptosporidium* in these three extra-alimentary cases described here was not identified. In other published reports of reptilian cryptosporidiosis, the taxonomic identification has been *C. serpentis*. One of the authors (FLF) has been employing an ELISA test (ProSpecT® *Cryptosporidium* EIA Assay, Alexon-Trend Inc., Ramsey, MN) for several years as part of a clinical trial to confirm the diagnosis of cryptosporidiosis in reptiles and amphibians, and he has found that the mammalian-based reagents employed in this test system cross-react with formalin-preserved reptilian fecal and tissue specimens.

As more attention is focused on the diseases of reptiles, it is likely that additional instances of extra-alimentary cryptosporidiosis will be disclosed. It is unfortunate that the origin of infection in each of these cases was not discovered, because it would have been valuable to know when and how these three lizards were infected. Furthermore, such such knowledge would help shed light upon the pathobiology of this important reptilian protozoan pathogen.

At the time of this writing, TKG is studying the tissues of all three of these lizards and trying to establish the speciation of the *Cryptosporidium* involved in each case. The results of these studies will be reported in a separate communication.
REFERENCES


<table>
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<tr>
<th>ACCESSION NUMBER</th>
<th>SPECIES</th>
<th>SEX</th>
<th>AGE</th>
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<td>97 LN-006</td>
<td><em>Iguana iguana</em></td>
<td>female</td>
<td>adult</td>
<td>Renal cryptosporidiosis; renal gout; peritubular interstitial and glomerular fibrosis; hepatocellular vacuolar lipidosis; medial arteriosclerosis; intraluminal intestinal ascaridoid helminthiasis; ulcerative enteritis.</td>
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<td>98 LN-037</td>
<td><em>Calumma parsoni cristifer</em></td>
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<td>Renal cryptosporidiosis; renal gout; interstitial fibrosis; pancreatic vacuolar lipidosis; necrotizing enterocolitis; aspiration pneumonia.</td>
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<td>V98-826</td>
<td><em>Iguana iguana</em></td>
<td>female</td>
<td>6 yr</td>
<td>Sialoadenitis associated with Cryptosporidium; tubuloalveolar epithelial hyperplasia; lymphoplasmacytic infiltration.</td>
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