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Source: Journal of Wildlife Diseases, 22(1) : 55-59

Published By: Wildlife Disease Association

URL: <https://doi.org/10.7589/0090-3558-22.1.55>

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## TYZZER'S DISEASE AS A COMPLICATION OF CANINE DISTEMPER IN A RACCOON

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**ABSTRACT:** Intercurrent canine distemper and Tyzzer's disease were diagnosed in a mature raccoon (*Procyon lotor*) submitted for necropsy. Clinical, gross and microscopic findings characteristic of canine distemper virus (CDV), included ataxia, dyspnea, suppurative conjunctivitis, interstitial pneumonitis and generalized lymphocytolysis. Inclusion bodies typical of CDV infection were present in many epithelial tissues. Acute multifocal hepatic necrosis and acute segmental necrotizing enteritis were attributed to the presence of *Bacillus piliformis* organisms in these lesions, confirmed by special stains and electron microscopy. This is apparently the first reported case of Tyzzer's disease in a raccoon.

### INTRODUCTION

Canine distemper has been described in a variety of carnivores (Budd, 1981; Blythe et al., 1983). Outbreaks of canine distemper in wild raccoons (*Procyon lotor*) have been reported from several localities in eastern and midwestern United States (Maurer and Nielsen, 1981) and as endemic disease in Ontario since 1981 (Cranfield et al., 1984).

Tyzzer's disease has been reported in many laboratory and domestic species including mice, rats, monkeys, hamsters, rabbits, cats, dogs and horses (Fujiwara, 1978). The disease is characterized by a triad of lesions including acute multifocal hepatic necrosis, segmental necrotizing enteritis and occasionally myocarditis. Tyzzer's disease in wild mammals was first reported in 1971 by Karstad et al. in muskrats (*Ondatra zibethica*), and it has been described subsequently in cottontail rabbits (*Sylvilagus floridanus*) (Ganaway et al., 1976), captive coyotes (*Canis latrans*) (Marler and Cook, 1976), snow leopards (*Panthera uncia*) (Schmidt et al., 1984) and gray fox (*Urocyon cinereoargenteus*) (Stanley et al., 1978).

This report describes the natural occur-

rence of Tyzzer's disease in a raccoon with canine distemper.

### MATERIALS AND METHODS

On 17 October 1983 an adult raccoon presented in a moribund state to a local humane society was submitted to the Department of Pathology at the Ontario Veterinary College for necropsy as a rabies suspect. Prior to death the animal had shown signs of ataxia, dyspnea and crusting around the eyes, suggestive of canine distemper. At necropsy, the animal was examined grossly and various specimens were collected in 10% buffered neutral formalin. Tissues were processed for histology, sectioned at 6  $\mu$ m thickness and stained with hematoxylin and eosin, Warthin-Starry, and Brown and Brenn stains. Formalin-fixed tissues were processed also for electron microscopy. Part of the brain was sent to the Rabies Diagnostic Laboratory, Agriculture Canada for rabies examination. Liver and spleen were submitted for aerobic bacterial culture.

### RESULTS

**Gross findings:** At necropsy, the animal was in poor body condition. The areas around the eyes were bilaterally crusted and the eyes were sunken, consistent with a state of marked dehydration. The hair coat was rough, and numerous fleas were present. There was focal epidermal ulceration and myiasis in the perineal area. Internally, there was abundant subcutaneous and abdominal fat. The lungs were moderately congested with patchy areas

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Received for publication 20 May 1985.

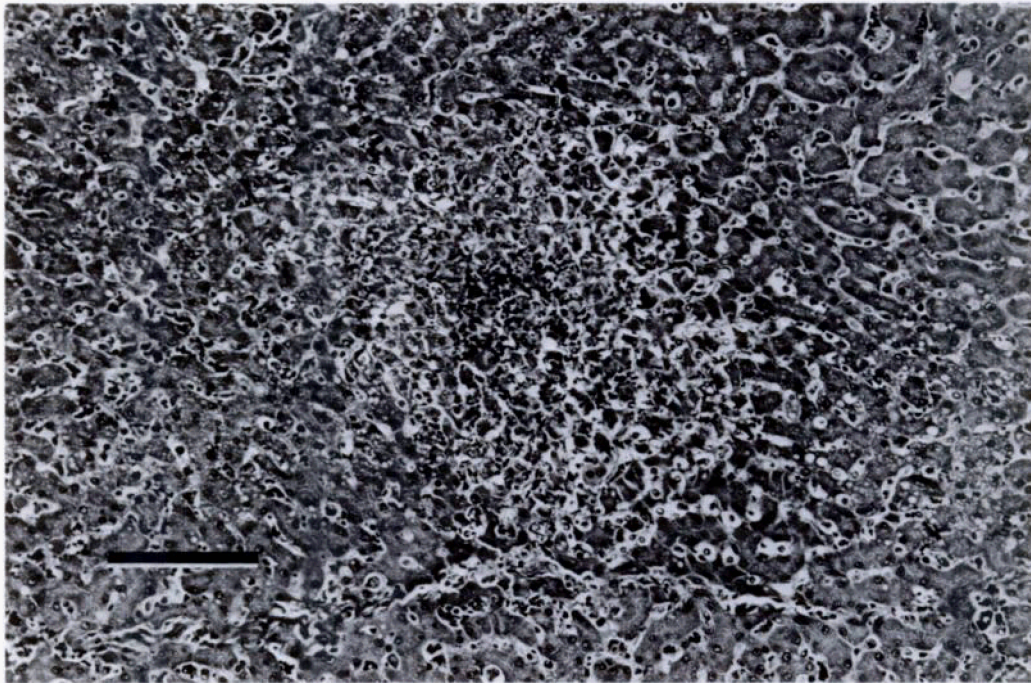


FIGURE 1. Focus of acute necrosis in the liver of a raccoon. H&E stain. Scale bar = 200  $\mu$ m.

of atelectasis and a pale consolidated area approximately 1 cm in diameter was present in the left diaphragmatic lung lobe. The liver was enlarged with numerous white foci up to 2–3 mm in diameter on the surface and throughout the parenchyma. The foci varied in appearance from white in smaller lesions to red with a white rim in larger foci. The spleen was enlarged and prominent lymphoid follicles were evident on cut surfaces. The gastrointestinal tract was devoid of ingesta, containing only bile-stained mucoid material. The ileum appeared congested, but otherwise was unremarkable grossly. Mesenteric lymph nodes were enlarged and congested.

**Microscopic findings:** The lungs were moderately congested with patchy areas of atelectasis. The single large focus of consolidation had many large neutrophils, and fewer macrophages and eosinophils, filling the alveolar spaces. Many of the in-

filtrating cells were necrotic and there were several foci of hemorrhage within this area. In less severely affected portions of lung there were scattered alveolar macrophages and occasional eosinophils. Bronchi and bronchioles contained sloughed epithelium and cellular debris. Eosinophilic cytoplasmic inclusion bodies were present in bronchiolar epithelial cells, biliary epithelium, pancreatic ductal epithelium, the urothelium of the renal pelvis, the transitional epithelium of the urinary bladder, the mucous neck cells of the gastric mucosa and follicular epithelial cells of the thyroid, as well as within syncytial cells present in the colloid of thyroid follicles.

Periarteriolar lymphoid aggregates in the spleen were markedly hypocellular. Cortical follicles in lymph nodes were depleted similarly. Numerous eosinophils and macrophages were present in the medullary sinuses. Foci of bacteria and

hemorrhage also were randomly scattered in the medulla. There were no visible lesions in the brain.

In the liver, there were numerous randomly distributed foci of acute coagulation necrosis (Fig. 1), many of which contained central hemorrhage. There was a minimal inflammatory reaction, consisting largely of eosinophils, associated with these lesions. Groups of elongate basophilic rods were identified within hepatocytes around the periphery of these areas. The characteristic "straw broom" clumped appearance of Tyzzer's organisms (*Bacillus piliformis*) was better demonstrated by Warthin-Starry silver stain (Fig. 2).

In the ileum, there were segmental areas of loss of crypts, with dilation of some crypts which were lined by flattened epithelium (Fig. 3). The epithelium lining other crypts was necrotic and the lumen often contained cellular debris. There was a marked eosinophilic and mononuclear leukocytic infiltrate in the lamina propria and subtending submucosa. Groups of bacterial rods, resembling those found in the liver, were demonstrated by Warthin-Starry silver stain in enterocytes lining the crypts.

**Bacteriology:** Aerobic culture of liver on blood agar and MacConkey's agar revealed the presence of moderate numbers of *E. coli* and a few *Pseudomonas aeruginosa*. Moderate numbers of untypable Group C *Salmonella* (6,7:K:-) and a few *Pseudomonas aeruginosa* were cultured from the spleen.

**Virology:** The caudal quadrant of the brain was submitted for rabies virus determination by fluorescent antibody and mouse inoculation techniques. Results were negative.

**Electron microscopy:** Follicular cells of the thyroid contained numerous viral aggregates within the cytoplasm, typical of canine distemper virus (Confer et al., 1975).

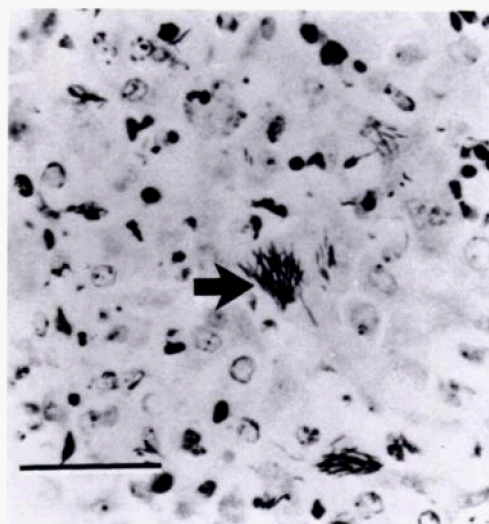


FIGURE 2. Numerous Tyzzer's bacilli (arrow) in hepatocytes at the periphery of a focus of necrosis in the liver of a raccoon. Warthin-Starry stain. Scale bar = 50  $\mu$ m.

Hepatocytes from the periphery of the focal areas of necrosis in the liver had numerous electron-dense organisms free in the cytoplasm. Viewed on cross-section, these bacteria had a dense granular core separated artefactually from the surrounding thick electron-dense membrane by a clear space. Occasional organisms had small clear areas within the dense core. The bacilli measured 0.53  $\mu$ m to 1.20  $\mu$ m in diameter. Other ultrastructural changes within the cell were attributed to a combination of cellular degeneration and postmortem change.

#### DISCUSSION

The finding of intracytoplasmic eosinophilic inclusion bodies in epithelial cells from a variety of tissues is characteristic of canine distemper. It has been hypothesized that canine distemper may facilitate intercurrent infections by its anergic or immunosuppressive effects, likely due to lymphocyte destruction (Mangi et al., 1976; Olsen and Krakowka, 1984). There are numerous reports of canine distemper,





FIGURE 3. Cryptal necrosis (arrow) and loss of crypts (arrowhead) in the ileum of a raccoon. H&E stain. Scale bar = 200  $\mu$ m.

in species other than raccoons, occurring concurrently with infections such as toxoplasmosis (Moller and Nielsen, 1964), listeriosis (Jakowski and Wyand, 1971), herpesvirus (Diters and Nielsen, 1978), cryptosporidiosis (Fukushima and Helman, 1984) and Tyzzer's disease (Poonacha and Smith, 1976). Cryptosporidiosis was reported previously in 42% of wild raccoons with canine distemper at the Metropolitan Toronto Zoo (Cranfield et al., 1984). In 1983, eight out of 57 raccoons submitted to the Department of Pathology, Ontario Veterinary College with confirmed or presumptive canine distemper, also had a concurrent infection with either cryptosporidiosis, toxoplasmosis, salmonellosis or some other bacterial disease (Necropsy Records, Wildlife Diseases Section, Department of Pathology, Ontario Veterinary College).

This case of Tyzzer's disease, the first

reported to our knowledge in a raccoon, extends the range of infections known to occur concurrently with canine distemper in that species. Its presence in an animal probably immunocompromised by canine distemper is consistent with the suggestion that Tyzzer's disease most commonly occurs in stressed or immunosuppressed hosts (Wobeser, 1981). Infection by *E. coli*, *Salmonella* and *Pseudomonas aeruginosa* likely represents a terminal event in this case.

#### ACKNOWLEDGMENTS

This study was supported by the Ontario Ministry of Natural Resources.

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