BioOne COMPLETE

Cryptosporidiosis in a Cotton Rat (Sigmodon hispidus)

Authors: Elangbam, Chandikumar S., Quails, Charles W., Ewing, Sidney A., and Lochmiller, Robert L.

Source: Journal of Wildlife Diseases, 29(1) : 161-164

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-29.1.161

BioOne Complete (complete.BioOne.org) is a full-text database of 200 subscribed and open-access titles in the biological, ecological, and environmental sciences published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Complete website, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/terms-of-use.

Usage of BioOne Complete content is strictly limited to personal, educational, and non - commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

BioOne sees sustainable scholarly publishing as an inherently collaborative enterprise connecting authors, nonprofit publishers, academic institutions, research libraries, and research funders in the common goal of maximizing access to critical research.

Downloaded From: https://bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://bioone.org/terms-of-use

Journal of Wildlife Diseases, 29(1), 1993, pp. 161–164
© Wildlife Disease Association 1993

Cryptosporidiosis in a Cotton Rat (*Sigmodon hispidus*)

Chandikumar S. Elangbam,[1,4] Charles W. Qualls., Jr.,[1] Sidney A. Ewing,[2] and Robert L. Lochmiller,[3] [1] Department of Veterinary Pathology, College of Veterinary Medicine, Stillwater, Oklahoma 74078, USA; [2] Department of Parasitology, College of Veterinary Medicine, Stillwater, Oklahoma 74078, USA; [3] Department of Zoology, Oklahoma State University, Stillwater, Oklahoma 74078, USA. [4] Present address: WIL Research Laboratories Inc., 1407 Montgomery Township Road #805, Ashland, Ohio 44805, USA

ABSTRACT: Cryptosporidiosis, previously unreported in cotton rats (*Sigmodon hispidus*), was observed in one of nine cotton rats from Pryor, Oklahoma (USA). Infection was confined to the large intestine. Microscopically, numerous cryptosporidians measuring 2 to 3 μm in diameter were intimately associated with the luminal surface of colonic and rectal mucosae. The affected lamina propria of the large intestine was diffusely infiltrated by eosinophils, lymphocytes and macrophages. Ultrastructurally, numerous trophozoites and a single schizont were observed. Microvilli were displaced by the presence of cryptosporidians at the attachment site.

Key words: Cryptosporidiosis, ultrastructure, pathology, cotton rats, *Sigmodon hispidus*.

Cryptosporidiosis, a coccidial parasite disease caused by various *Cryptosporidium* spp. has been recognized in several mammalian, reptilian and avian species (Angus, 1983; Davis and Jenkins, 986; Fayer and Ungar, 1986). Because of clinical disease in humans with acquired immune deficiency syndrome, the disease has gained increased interest. Diarrhea has been associated with infection in lambs, calves, pigs, rhesus monkeys, mice, rats, turkey poults and humans (Fox et al., 1984; Angus et al., 1985; Aboul-Magd et al., 1989; Ungar et al., 1990; Moody et al., 1991). Lethargy, rough hair coat, failure to gain or maintain weight, and weight loss have been reported in natural and experimental infections of mice, rats and rabbits (Harkness and Wagner, 1989). Furthermore, a progressive infection, fatal within 4 mo with weight loss and intermittent diarrhea has been reported in adult athymic mice (Ungar et al., 1990) and immunocompromised rats (Aboul-Magd et al., 1989). A lowered mucosal enzyme activity of the small intestine has been associated with cryptosporidiosis in guinea pig and calves (Manning et al., 1984; Barker and Dreumel, 1985).

Cryptosporidium spp. develops in the absorptive surface of intestinal epithelial cells and is associated with villus atrophy of varying severity, blunting and fusion of villi, and hypertrophy of crypts of Lieberkuhn (Gibson and Wagner, 1986; Harkness and Wagner, 1989; Moody et al., 1991). Cryptosporidial infection is associated with loss and displacement of the enterocyte microvilli by the parasite (Van Kruiningen, 1988). Initially, a pentalaminar membrane fusion site develops at the base of the protozoa between the parasite's outer plasma membrane and host membrane. Later, the host membrane disintegrates and directly contacts absorptive cell cytoplasm (Harkness and Wagner, 1989; Marcial and Madara, 1986).

No report of cryptosporidiosis in cotton rats (*Sigmodon hispidus*) has been found in the literature. We describe cryptosporidiosis in a male cotton rat.

Nine cotton rats were collected from typical ungrazed, tall grass prairie in Pryor, Oklahoma (95°17′N, 36°15′W). Animals were returned to the laboratory, fasted overnight, humanely killed by cervical dislocation, and necropsied. Liver, kidneys, lungs, heart, spleen, stomach, duodenum, jejunum, ileum, colon, cecum, rectum, brain and muscle samples were fixed in Carson's modified Milloning's phosphate buffered formalin (Sheehan and Hrapchak, 1980). Six-micron thick sections of paraffin embedded tissue were stained with hematoxylin and eosin (H&E) and permanently mounted on glass slides for microscopic examination. Formalin-fixed tissues (1 mm cube) of colon and rectum were post-fixed for 2 hr in Karnovsky's fixative (Karnovsky, 1965) and prepared for transmission electron microscopy. Fif-

Downloaded From: https://bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://bioone.org/terms-of-use

ty-five nm sections of each sample were cut with glass knives on a Sorvall MT 6000 Ultracut ultramicrotome (Research Manufacturing Company, Phoenix, Arizona, USA), mounted on glass microscope slides, and stained with Mallory's stain (Richardson et al., 1960). Areas containing *Cryptosporidium* spp. were selected for thin-sectioning at 0.55 nm, stained with uranyl acetate and lead citrate (Pillsdury, 1980), and examined with a Jeol 100C transmission electron microscope (Jeol Ltd., Tokyo, Japan).

One of nine cotton rats had *Cryptosporidium* spp. in the large intestine with no recognized clinical signs. The liver was fragile, pale and enlarged with accentuated lobular pattern. All other organs (lung, kidneys, heart, stomach, small intestine, brain, muscle samples and genital organs) appeared normal. There was a moderate enlargement of the centrilobular and midzonal hepatocytes in the liver, including enlarged nuclei and increased cytoplasmic vacuolations; there also were multifocal aggregates of lymphocytes and macrophages. No *Cryptosporidium* spp. was observed in the liver.

Macroscopically, the intestinal mucosa and contents of this *Cryptosporidium* spp.-infected cotton rat were normal. Microscopically, the colonic and rectal mucosa were diffusely infiltrated by mixed inflammatory cells (eosinophils, lymphocytes, macrophages). No such lesions were observed in the esophagus, stomach, duodenum, jejunum and ileum. Rarely, focal necrosis of superficial mucosa and ectatic crypts containing cellular debris and neutrophils were present in the large intestine. Numerous cryptosporidian trophozoites were intimately associated with the luminal surface of the epithelium of the large intestine; most were approximately 2 to 3 μm in diameter and were easily seen in sections stained with hematoxylin and eosin.

Based on transmission electron microscopy, numerous cryptosporidians occurred at the luminal border of epithelial cells in the large intestine. The microvilli of epithelial cells were displaced by the presence of parasites at the attachment site. Numerous trophozoites and a single schizont were the only parasite stages observed in the section (Figs. 1, 2). Trophozoites were variable in shape and measured 1.99 to 2.39 μm by 2.39 to 3.31 μm; they were recognized by the presence of a nucleus with a nucleolus, endoplasmic reticulum, and Golgi anlagen (Vetterling et al., 1971). Trophozoites were attached to the apical surface of enterocytes. There were broad, electron-dense attachment zones at the interface between parasite and host cells. The schizont measured 2.70 by 3.39 μm; it was identified by the presence of membrane-bound merozoites free within the parasitophorous vacuole. These ultrastructural features are characteristic of *Cryptosporidium* spp., as described by Vetterling et al. (1971).

In laboratory animals (guinea pigs, rats, mice, rabbits, ferrets), developmental stages of *Cryptosporidium* spp. may be seen from the pylorus or duodenum to the cecum with the highest concentration in the ileum (Rehg et al., 1988; Harkness and Wagner, 1989). The small intestine was not affected in this cotton rat. It is not known whether the mild eosinophilic inflammation of colon and rectum in the absence of helminth parasites, that we observed in this cotton rat was associated with *Cryptosporidium* spp. infection. However, similar observations have been reported in ferrets (Rehg et al., 1988).

Six of the nine cotton rats, including the one with cryptosporidiosis, were collected from a site contaminated with Aroclor 1254, a polychlorinated biphenyls (PCB) mixture; the other rats were collected from an uncomtaminated site with ecologically similar habitat, approximately 0.8 km from the contaminated site. The soil concentration of Aroclor 1254 was more than 800 ppm in some areas (United States Corps of Engineers, 1987). Tissue PCB concen-

Downloaded From: https://bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://bioone.org/terms-of-use

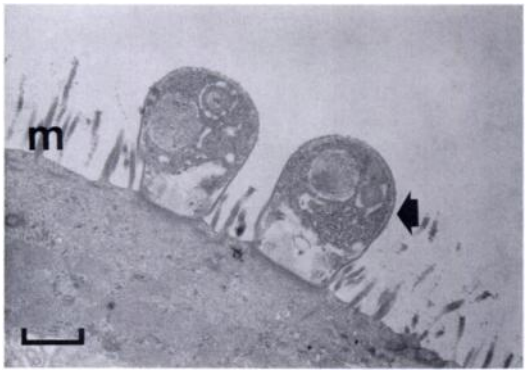


FIGURE 1. Electron micrograph of cotton rat intestine with two trophozoites (arrow) attached to the epithelial cell and displacing microvilli (m). Uranyl acetate and lead citrate. Scale bar = 1 μm.

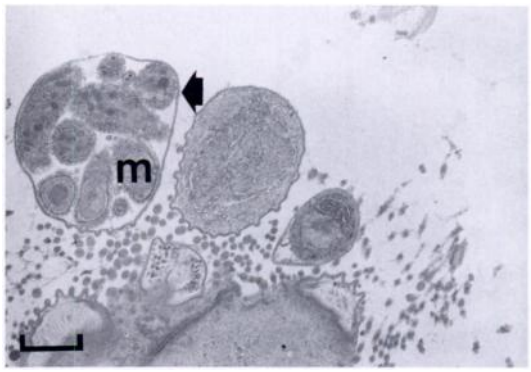


FIGURE 2. Electron micrograph of a schizont (arrow) containing merozoites (m) separated by artifact from an epithelium. Uranyl acetate and lead citrate. Scale bar = 1 μm.

trations of contaminated cotton rats (1.42 to 12.36 ppm) greatly exceeded the concentrations in uncontaminated rats (<0.05 ppm). Hepatic changes in this infected cotton rat were similar to those seen in the remaining contaminated rats and probably were related to the Aroclor 1254 exposure. The hypertrophy of hepatocytes with increased organelle content has been reported in PCB-fed rats and rabbits (Sleight and Sanger, 1976; Raber and Carter, 1986). Others (Cooper et al., 1984; Fukushima and Helman, 1984) suggest that *Cryptosporidium* spp. acts as an opportunist in immunocompromised hosts. In another study, cryptosporidial infection was induced in cyclophosphamide-treated immunosuppressed rats which remained infected as long as they received cyclophosphamide (Rehg et al., 1987). It is possible that exposure to Aroclor 1254, a known immunotoxin, predisposed this cotton rat to cryptosporidiosis. However, we did not evaluate the immunological status of the host and, therefore, cannot confirm immunodeficiency nor speculate further on its role in the development of cryptosporidiosis.

The source of infection in this cotton rat is not known. Interspecies transmission of *Cryptosporidium* spp. is well documented (Tzipori et al., 1980; Iseki et al., 1989), but no attempt was made in this instance to determine the source of infection.

LITERATURE CITED

ABOUL-MAGD, L. A., A. M. EL-RIDI, AND S. A. MICHAEL. 1989. Biological and hematological changes in experimental cryptosporidiasis. Journal of the Egyptian Society of Parsitology 19: 49–56.

ANGUS, K. W. 1983. Cryptosporidiosis in man, domestic animals and birds: Review. Journal of the Royal Society of Medicine 76: 62–69.

———, G. HUTCHISON, AND H. M. MUNRO. 1985. Infectivity of a strain of cryptosporidium found in the guinea pig (*Cavia percellus*) for guinea pigs, mice and lambs. Journal of Comparative Pathology 95: 151–165.

BARKER, I. K., AND V. A. A. DREUMEL. 1985. The alimentary system. *In* Pathology of domestic animals, K. V. F. Jub, P. C. Kennedy, and N. Palmer (eds.). Academic Press Inc., New York, New York pp. 1–237.

COOPER, D. A., A. WODAK, D. J. E. MARRIOT, J. L. HARKNESS, M. RALSTON, A. HILL, AND R. PENNY. 1984. Cryptosporidiosis in the acquired immune deficiency syndrome. Pathology 16: 455.

DAVIS, A. J., AND S. J. JENKINS. 1986. Cryptosporidiasis and proliferative ileitis in a hamster. Veterinary Pathology 23: 632–633.

FAYER, R., AND B. L. P. UNGAR. 1986. *Cryptosporidium* spp. and cryptosporidiosis. Microbiological Reviews 50: 458–483.

FOX, J. G., C. E. NEWCOMER, AND H. ROZMIAREK. 1984. Selected zoonoses and other health hazards. *In* Laboratory animal medicine, J. G. Fox, B. J. Cohen, and D. Loew (eds.). Academic Press Inc., Orlando, Florida, pp. 613–648.

FUKUSHIMA, K., AND R. G. HELMAN. 1984. Cryp-

Downloaded From: https://bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://bioone.org/terms-of-use

tosporidiasis in a pup with distemper. Veterinary Pathology 21: 247.

GIBSON, S. V., AND J. E. WAGNER. 1986. Cryptosporidiosis in guinea pigs: A retrospective study. Journal of American Veterinary Medical Association 189: 1033–1034.

HARKNESS, J. E., AND J. E. WAGNER. 1989. The biology and medicine of rabbits and rodents. Lea & Febiger, Philadelphia, Pennsylvania, pp. 129–131.

ISEKI, M., T. MAEKAWA, K. MORIYA, S. UNI, AND S. TAKADA. 1989. Infectivity of *Cryptosporidium muris* (strain RN 66) in various laboratory animals. Parasitology Research 75: 218–222.

KARNOVSKY, M. J. 1965. A formaldehyde-glutaraldehyde fixative of high osmolality for use in electron microscopy. The Journal of Cell Biology 27: 137A.

MANNING, P. J., J. E. WAGNER, AND J. E. HARKNESS. 1984. Biology and diseases of guinea pigs. *In* Laboratory animal medicine, J. G. Fox, B. J. Cohen, and D. Loew (eds.). Academic Press Inc., Orlando, Florida, pp. 149–181.

MARCIAL, M. A., AND J. L. MADARA. 1986. Cryptosporidium: Cellular localization, structural analysis of absorptive cell–parasite membrane–membrane interactions in guinea pigs, and suggestion of protozoan transport by M cells. Gastroenterology 90: 583–594.

MOODY, K. D., D. G. BROWNSTEIN, AND E. A. JOHNSON. 1991. Cryptosporidiosis in suckling laboratory rats. Laboratory Animal Science 41: 625–627.

PILLSDURY, N. 1980. Electron microscopy. *In* Theory and practice of histotechnology, D. C. Sheehan and B. B. Hrapchak (eds.). Battelle Press, Columbus, Ohio, pp. 327–346.

RABER, B. T., AND J. W. CARTER. 1986. Localization of ultrastructural alterations induced in rat liver by dietary polybromobiphenyls (FireMaster BP-6). Archives of Environmental Contamination and Toxicology 15: 725–732.

REHG, J. E., M. L. HANCOCK, AND D. B. WOODMANSEE. 1987. Characterization of cyclophosphamide-rat model of cryptosporidiosis. Infection and Immunity 55: 2669–2674.

———, F. GIGLIOTTI, AND D. C. STOKES. 1988. Cryptosporidiosis in ferrets. Laboratory Animal Science 38: 155–158.

RICHARDSON, K. C., L. JARETT, AND E. H. FINKE. 1960. Embedding in epoxy resin for ultrathin sectioning in electron microscopy. Stain Technology 35: 313–325.

SHEEHAN, D. C., AND B. B. HRAPCHAK. 1980. Fixation. *In* Theory and practice of histotechnology. Battelle Press, Columbus, Ohio, pp. 41–58.

SLEIGHT, S. D., AND V. L. SANGER. 1976. Pathologic feature of polybrominated biphenyls (PBB) toxicosis in the rat and guinea pig. Journal of American Veterinary Medical Association 169: 1231–1235.

TZIPORI, S., K. W. ANGUS, I. CAMPBELL, AND E. W. GRAY. 1980. *Cryptosporidium*: Evidence for a single-species genus. Infection and Immunity 30: 884–886.

UNGAR, B. L., J. A. BURRIS, C. A. QUINN, AND F. D. FINKELMAN. 1990. New mouse models for chronic *Cryptosporidium* infection in immunodeficient hosts. Infection and Immunity 58: 961–969.

UNITED STATES CORPS OF ENGINEERS. 1987. Fort Gibson Lake, Oklahoma, Phase I—Remedial investigation/feasibility study, United States Corps of Engineers, Tulsa, Oklahoma, pp. 1–4.

VAN KRUININGEN, H. J. 1988. Gastrointestinal system. *In* Special veterinary pathology, R. G. Thomson (ed.). B. C. Decker Inc., Philadelphia, Pennsylvania, pp. 133–227.

VETTERLING, J. M., A. TAKEUCHI, AND P. A. MADDEN. 1971. *Cryptosporidium wrairi* from the guinea pig. The Journal of Protozoology 18: 248–260.

Received for publication 9 December 1991.

Downloaded From: https://bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://bioone.org/terms-of-use