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EXPERIMENTAL INFECTION OF DOGS WITH *Sarcocystis* FROM WAPITI [1]

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Abstract: Ten domestic dogs became infected with *Sarcocystis* when fed single portions of heart, esophagus and diaphragm from a two-year-old female wapiti *(Cervus canadensis).* The prepatent period was 14 days in all exposed dogs; the patent period ranged from 8 to 20 days. Neither the 10 control dogs, nor two dogs fed sporocysts collected from the infected dogs passed sporocysts within the study period. Sporocysts averaged 16.5 by 11.1 μm in size.

INTRODUCTION

Sarcocystis is a common parasite of many wild and domestic animals.[2] The life cycles of the species thus far studied are of the predator-prey type, with gametogony in intestinal cells of a carnivore/predator and merogony in endothelial, muscle and nerve cells of the herbivore/prey.[9] Some of the species for which the life cycle is known are common in cattle *(Bos taurus),*[4] sheep *(Ovis aries)*[10] and mule deer *(Odocoileus hemionus).*[6,7]

Neither the exogenous nor the endogenous stages of the *Sarcocystis* sp. infecting wapiti *(Cervus canadensis)* have been studied, although the parasite is common in these animals.[1,5,8,11,12,13] This study was conducted to determine if the dog is a suitable definitive host for a *Sarcocystis* sp. from wapiti and, if so, to describe the sporocysts and compare them to those already identified and characterized from carnivores infected with species from the tissues of other ruminants.

MATERIALS AND METHODS

Seven male and 13 female purebred or mixed-breed dogs, ranging from six months to about eight years of age were used in the study. They were purchased from an animal control facility, transported to the research facility, treated with anthelmintics, and given routine vaccinations. They were housed in groups of two or three in 3.10 × 1.55 m steel kennels on concrete floors and provided with a dry commercial dog food and water *ad libitum.*

A fecal sample from each dog was collected and examined upon arrival, three times each week until the test dogs were infected, and daily thereafter until sporocysts were not found in any stool specimens for 14 consecutive days. Two grams of each fecal specimen were mixed with tap water, an equal volume of Sheather's sucrose solution was added, mixed, and the suspension was centrifuged at 250 × g. A bacteriologic loop or a coverslip was used to transfer a film of solution from the top of the flotation medium to a glass slide, which was then examined microscopically for sporocysts. Total fecal output was collected from dogs with patent infections, the sporocysts concentrated by sucrose flotation, and stored in tap water at 8 C. Ten sporocysts selected from specimens pooled daily were measured on the sec-

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ond day of the patent period and each third day thereafter until a total of 50 sporocysts had been measured. The sporocysts randomly selected for measurement and photography were collected, concentrated, pooled, measured and photographed within 24 h after the source stools were passed.

The heart, esophagus and diaphragm from a two-year-old wapiti female were collected at slaughter, packed in ice while in transit to the laboratory, examined microscopically at 20 × to ascertain the presence of sarcocysts, and stored for three days at 4 C. The chilled tissues were diced, mixed and divided into ten, 340 g portions. Each of 10 dogs, randomly selected from the group of 20, was given one of the tissue portions in a single feeding. After each dog had consumed its entire portion of infective tissue, it was penned with one or two of its infected group mates in one of four pens. The 10 control dogs received only the dry food throughout the study period.

Two control dogs, about 6 and 12 months old, respectively, were each fed 100,000 freshly collected sporocysts *per os* to determine if sporocysts would initiate gametogeny. Their feces were examined for sporocysts daily for 25 days.

Seven dogs fed the infected tissue and five control dogs were killed during the prepatent period for collection of tissues for another study. Three dogs remained in the test group and five in the control group when the patent period began and tissue collection was terminated.

RESULTS

All three dogs fed infected tissue and remaining alive throughout the experiment began to pass sporocysts in their feces on the 14th day after infection. The sporocysts ranged in size from 18.7 to 14.0 by 13.4 to 9.5 μm (average 16.5 by 11.1 μm) and lacked Stieda bodies (Fig. 1). On the first day of patency, sporocysts comprising less than 1% of the pooled specimens were seen in the oocyst form,

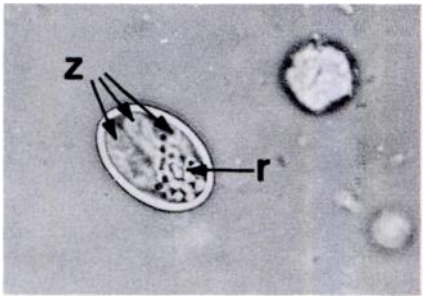

FIGURE 1. Sporocyst showing residual body (r) and 3 of 4 sporozoites (z).

the wall of which appeared membranous and without a micropyle (Fig. 2). The dogs passed sporocysts for 8 successive days, 14 successive days and 20 successive days, respectively. One of the infected dogs passed a mixture of two sizes of unsporulated isosporan oocysts seven and eight days after eating the infected meat; one oocyst type averaged 37.0 by 31.5 μm whereas the other averaged 12.0 by 10.0 μm in size. With that exception, no coccidian cysts of any kind were seen in the fecal flotations from any specimens from the time the dogs arrived until the patent period began in the three dogs experimentally infected. None of the control dogs passed sporocysts or oocysts during the study period, nor were any coccidian tissue stages seen in those examined at necropsy. Neither of the two dogs fed sporocysts passed sporocysts during the 25 days their feces were examined. No clinical signs of coccidiosis were seen in infected or control dogs during the study.

DISCUSSION

This study established the suitability of the domestic dog as a definitive host for a species of *Sarcocystis* found in wapiti. Other carnivores in addition to domestic dogs serve as definitive hosts for species of *Sarcocystis* from herbivores other than wapiti,[2] but the role of other carnivores in this cycle remains to

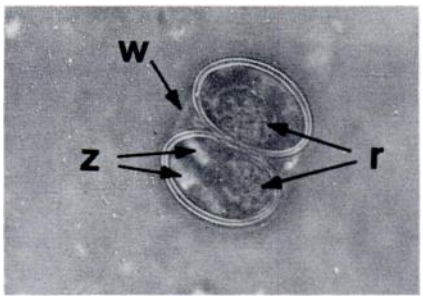

FIGURE 2. Oocyst showing 2 sporocysts enclosed by membranous oocyst wall (w).

be determined. The interaction of domestic dogs with wapiti is limited mainly to areas where a portion of a wapiti range nears farms, ranches or towns where dogs are allowed to roam free. One or more of many wild carnivorous mammals and birds, much more closely associated with wapiti than dogs, probably contribute to the high prevalence of *Sarcocystis* infection in wapiti. The prevalence of the parasite in Wyoming is greater than 90% in animals older than one year of age.[8]

The sporocysts of this species differ significantly (P=0.01) in size (16.5 × 11.1 μm) from those of mule deer-dog origin (14.5 × 9.2 μm),[6,7] sheep-dog origin (14.8 × 9.9 μm),[2,9] pig-dog origin (12.6 × 9.6 μm)[2,9] and horse-dog origin (15.2 × 10.0 μm)[2,9] but not from those of ox-dog origin (16.3 × 10.8 μm).[2,3,7,9] Despite the similarity in size of *S. cruzi* sporocysts to those from the wapiti-dog cycle they may be different species, since no species has been found that parasitizes more than one genus of intermediate host.[9] In addition, the prepatent period of *S. cruzi* in the dog was 9-22 days,[2,3] whereas that of the species from the wapiti in the dog was 14 days. Other studies are in progress to determine if they are separate species.

The dog which passed the isosporan oocysts prior to the appearance of sporocysts may have been infected with another coccidian prior to this study, it may have become infected by an infective tissue stage in the wapiti flesh, or sporulated oocysts may have gotten into the dry food or water by an unknown means. Certain isosporan species may utilize an intermediate host but not require one.[9] Had the isosporan been in the wapiti tissue, one or more of the other five dogs alive at that time and fed the infected flesh likely would have passed oocysts; had the dried food or water been contaminated with sporulated oocysts, any of the other test or control dogs could have passed oocysts. Had any of the dogs during the study acquired new, extraneous infections by any means, tissue stages in addition to those seen resulting from the experimental infection presumably would have appeared in the necropsy sections; none did. The oocysts may have resulted from an infection acquired prior to this study and were passed after an unknown physiological factor stimulated completion of development.

The patent periods in this study cannot be compared with those of others, since the dogs in other studies often received multiple feedings of infected meat. In addition, no satisfactory method has been described for quantitating the infective units in a given amount of tissue. Probably as a result of these factors, many previous studies, as well as this one, have shown inconsistent or variable prepatent or patent periods in the carnivores.[2,3,6,9]

The sporocysts of this species apparently will not initiate a patent infection in the dog, since neither clinical signs nor sporocysts resulted from inoculating the two young dogs with sporocysts. These results support those of Fayer,[3] although tissue sections from the dogs in our study were not examined as they were in his.

Of the *Sarcocystis* species studied, *S. cruzi* from the ox-dog cycle resembles the species of the wapiti-dog cycle more closely than the others. Sporocyst size and prepatent periods differ, although not

decisively enough to differentiate between the two. Intermediate host specificity, pathology, serology and excystation studies are in progress, and should help define the taxonomic status of this species.

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