Brief Communication

ENIGMATIC CYST-FORMING SPOROZOON IN THE SPINAL CORD OF A DOG

A female racing greyhound dog was presented to the clinic at the age of 3 months with a 2 months' history of irregular hindleg movements and thin hindleg muscles. On examination 1 month later, a squatting standing posture, a week proprioceptive positioning reaction, and severe atrophy and pain of the lumbar and thigh muscles, were evident. The dog was sacrificed and necropsied. The adrenals, hindleg muscles, kidneys, myocardium, peripheral nerves, spinal cord, thyroid, and tonsils were fixed in 10 % neutral buffered formalin and processed for histopathology.

Nonpurulent meningomyelitis, foci of gliosis and malacia were found thoughout the spinal cord. Radiculitis was also evident. Toxoplasma-like cysts (Fig. 1) were often seen in the spinal cord. Necrotic and regenerative inflammatory foci were common in the skeletal muscles and myocardium but microorganisms were not found there.

Paraffin embedded blocks of the spinal cord were reprocessed for electron microscopy as described previously (*Lindberg* 1986). Ultrathin sections were stained with uranyl acetate/lead citrate.

The subspherical cysts (Figs. 1 and 2) were about $10-20\times5$ $-10~\mu m$ in size. A cyst wall, $1-2.5~\mu m$ thick, was always present. Banana-shaped, oval or globular organisms about $4-6\times1-1.6~\mu m$, varying in number from 40 to 100, were present in each cyst.

A definite parasitophorous vacuole was present. The primary cyst wall was dense and finely granular. The substance between the organisms was less dense and there were no septa. The organisms (Fig. 3) showed a single nucleus, a three-layered pellicle and a conoid. 2—6 osmiophilic structures, apparently rhoptries, were observed. The subpellicular microtubules were 22 in number and 50 to 80 micronemes were present. Amylopectin granules were in the cytoplasm of the organism. Division

occurred by endodyogony resulting in 2 merozoites within a mother cell.

The ultrastructural characteristics of the organisms were compatible with those of motile stages of sporozoa (Chobotar & Scholtyseck 1982). Due to lack of unfixed material, experimental infection with the parasite was not performed. The occurrence of a cyst stage suggests a cyst-forming sporozoon, probably a coccidian. Cyst-forming coccidia include Toxoplasma, Hammondia, Sarcocystis, Frenkelia and Besnoitia (Dubey 1977).

No serologic test for toxoplasmosis was done on this case. However, immunoperoxidase staining (PAP/DAB) on tissue sections of the parasite was negative for Toxoplasma. Since the parasite also showed 50—80 micronemes, while Toxoplasma has only 30—50 (Sulzer et al. 1977), we excluded that these organisms could be Toxoplasma. Due to antigenic relationship between Toxoplasma and Hammondia (Araujo et al. 1984) the negative immunoperoxidase staining does not support that the parasite belongs to Hammondia. Sarcocystis and Frenkelia could be ruled out due to the absence of metrocytes. Enigmatic bodies, characteristic of Besnoitia (Scholtyseck et al. 1973), were not seen.

An unidentified cyst-forming sporozoon, occurring in the central nervous system and skeletal muscles, was reported from dogs in Norway (*Bjerkås et al.* 1984). The cysts described were larger (up to 60 µm) than those found in this case and sometimes they lacked a cyst wall. However, the fine structure of the organism reported here was on the whole similar to that described from Norway.

We conclude that the parasite may belong to cyst-forming sporozoa which could be coccidia. It was not possible to relate the organism to any of the known genera of cyst-forming coccidia.

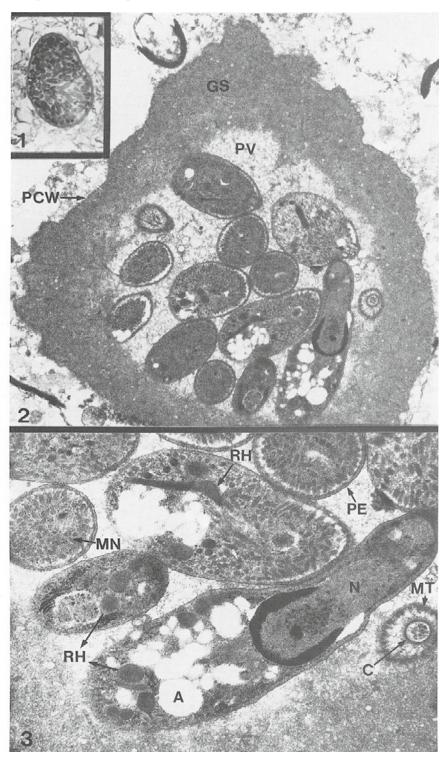
Figure 1. Semi-thin section of a cyst in the spinal cord. Toluidine blue \times 2500.

Figures 2, 3. Electron micrograph of a cyst, containing merozoites, in the spinal cord. Fig. 2. × 11700. Fig 3. × 25000.

Abbreviations:

A = Amylopectin; C = Conoid; GS = Ground substance; MN = Micronemes; MT = Microtubules; N = Nucleus; PCW = Primary cyst wall; PE = Pellicle; PV = Parasitophorous vacuole; RH = Rhoptries.

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