straight-chain primary alcohol, did not affect rice growth in three experiments. Compounds similar to triacontanol are present in other species (*9*) and may be active as plant growth regulators.

The response of crops to applications of triacontanol, alfalfa hay, and crude chloroform extracts of alfalfa must be compared in the field to directly associate the data reported here with the increased yields from small quantities of alfalfa that we have measured in the field.

STANLEY K. RIES, VIOLET WERT
Department of Horticulture, Michigan State University, East Lansing 48824
CHARLES C. SWEELEY
Department of Biochemistry, Michigan State University
RICHARD A. LEAVITT
Department of Entomology, Michigan State University

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Sarcocystosis: A Clinical Outbreak in Dairy Calves

Abstract. *Death and illness in a pen of eight yearling dairy heifers was caused by the protozoan parasite* Sarcocystis. *All animals had weight loss, weakness, marginal anemia, and elevated serum enzymes. Affected animals had high hemagglutinating antibody titers to* Sarcocystis *antigen. Affected tissues of the two animals that died demonstrated schizonts and young cysts during pathologic examination. The resident farm dog was shedding* Sarcocystis *sporocysts and was incriminated as the source of infection.*

Historically, the finding of *Sarcocystis* cysts in a high percentage of slaughtered domestic animals has been regarded with interest, but little concern has been shown because the parasite appeared nonpathogenic (*1*) and its life cycle was unknown except for the intramuscular cyst stage. *Sarcocystis* is a coccidian parasite (*2*); schizonts and cysts develop in domestic food animals (intermediate hosts) (*3, 4*) and gametes and sporocysts develop in carnivores (final hosts) (*5*). There have been several reports on the pathogenicity of some species of *Sarcocystis* for the intermediate host. Under experimental conditions the feeding of sporocysts from dogs to calves resulted in generalized clinical signs, including inappetence, weight loss, fever, anemia, and death (*3, 4, 6*). Schizonts or young cysts were seen in histologic sections from affected animals (*3, 4, 6*). Serum enzymes and antibody titers to *Sarcocystis* antigen were elevated (*7, 8*). Similarities were recognized between experimentally induced acute sarcocystosis and Dalmeny disease (*9–13*), an outbreak involving an unidentified protozoan organism in a herd of dairy cattle in Canada in 1961 (*14*). Recently it was postulated that numerous field cases of sarcocystosis may be unrecognized, resulting in considerable economic loss (*6*). Until now no documented field cases have been reported as acute sarcocystosis. Documentation of such a field case is presented in this report.

A dead heifer, from a pen containing eight yearling Holstein heifers on a dairy farm in Seneca County, central New York State, was presented to the Necropsy Service at the New York State College of Veterinary Medicine. The animal had been weak for 2 days and unable to rise the day before death. At necropsy, the carcass was emaciated and icteric, with an excessive amount of yellow fluid in the peritoneal and pleural cavities. The histologic findings included pneumonitis and mild leptomeningitis, with schizonts present in the vascular endothelium of many soft tissues (Fig. 1A). Death was attributed to a diffuse infection by a *Sarcocystis*-like organism.

A second heifer from the same pen was clinically examined at the Large Animal Hospital of the College. It was significantly underweight for its age, depressed, weak, and had a poor appetite. Temperature, pulse, and respiratory rates were within normal limits. Palpable lymph nodes were enlarged and there was obvious pallor of the visible mucous membranes. Hematologic evaluation and a bone marrow biopsy indicated the presence of a macrocytic hypochromic responsive anemia (hematocrit, 16 percent). Serum enzymes were elevated [lactate dehydrogenase (LDH), 451 I.U. and glutamic-oxaloacetic transaminase (GOT), 215 I.U.]. The serum albumin level was below normal (1.8 g/dl). Young cysts found in a cervical muscle biopsy provided strong presumptive evidence that the clinical signs were attributable to acute sarcocystosis. Serologically, the heifer was negative by the indirect hemagglutination (IHA) test for *Toxoplasma gondii*, a protozoan parasite related to *Sarcocystis*.

The heifer became moribund after 3 days of hospitalization, was killed, and a complete postmortem examination was performed. The carcass was emaciated and the mucous membranes were pale. All lymph nodes were moderately enlarged and edematous. Histologic findings included pneumonitis, diffuse degenerative myositis, and lymphoid hyperplasia. Young *Sarcocystis* cysts were found in all striated muscles examined.

An epidemiologic evaluation provided additional circumstantial evidence that the outbreak was acute sarcocystosis. Adjacent to the pen in which the sick heifers were housed, a dog had been tied for 3 weeks while it was in estrus. A fecal sample from this dog contained typical *Sarcocystis* sporocysts with four sporozoites and a granular residual body (Fig. 1B). Four weeks later, old dog feces found in the barn contained sporocysts, although fresh fecal material from the dog was negative. The dog had been fed three cow heads over the preceding 2 months; the last feeding was approximately 3 weeks before the death of the first heifer.

Intramuscular cysts found in the moribund heifer were examined by electron microscopy and contained only metrocytes (*15*). (Fig. 1C). After the death of this heifer, refrigerated meat from the carcass was fed to a dog and a cat for four consecutive days. Fecal samples were negative for *Sarcocystis* sporocysts from 2 days before feeding until 40 days after feeding. The electron microscopy and feeding trials suggest that the cysts in this heifer were immature and noninfectious.

The remaining six heifers on the farm were evaluated clinically, hematologically, and serologically. One heifer was obviously underweight for its age and had enlarged lymph nodes and pale mucous membranes. Two other heifers also had peripheral lymphadenopathy. The six animals had marginal anemia (mean hematocrit, 28 percent; range, 25 to 30 percent)

and elevated serum GOT (mean, 278 I.U.; range, 203 to 344 I.U.), and five had elevated serum LDH (mean, 438 I.U.; range, 372 to 473 I.U.). The clinical and hematologic findings in these natural cases of sarcocystosis closely resemble those reported in the experimental disease (*6, 7*).

Confirmation of *Sarcocystis* as the agent responsible for the clinical outbreak on the dairy farm was obtained by comparison of antibody titers from the killed heifer, the six affected heifers, and 19 other geographically isolated cows and calves on the premises (Fig. 2). Indirect hemagglutination tests were performed to determine circulating antibody titers to *Sarcocystis* antigen (*8*). The levels of antibody present in the killed heifer and the six affected heifers are comparable to those found in experimentally infected calves (*8*). The low titers of the 3-month-old calves indicate little or no prior exposure to *Sarcocystis* antigen. Titers of eight of the milking cows are comparable to titers previously found in normal healthy dairy cows (*8*). The elevated titers found in two cows indicate previous exposure to *Sarcocystis*, although no signs of clinical illness were noted during the current outbreak.

In summary, the clinical and hematologic findings and histologic demonstration of schizonts in the vascular endothelium aided in the recognition of a *Sarcocystis*-like infection. Serologic findings of markedly elevated titers to *Sarcocystis* antigen in the affected heifers verified the diagnosis as a natural outbreak of acute sarcocystosis, which until now has been unrecognized as a natural disease. Epidemiologically, the dog was strongly incriminated as the source of infection, although other carnivores cannot be excluded as final hosts (*16*).

This outbreak should focus attention on the economic impact of acute sarcocystosis causing death in food-producing animals, and on the economic loss due to poor growth rates. The latter may be of greater importance to food producers than the more spectacular clinical disease syndrome. As shown by these studies, serologic evidence is necessary to verify acute sarcocystosis, since the presence of protozoan organisms in tissues is suggestive of *Sarcocystis* but not definitive.

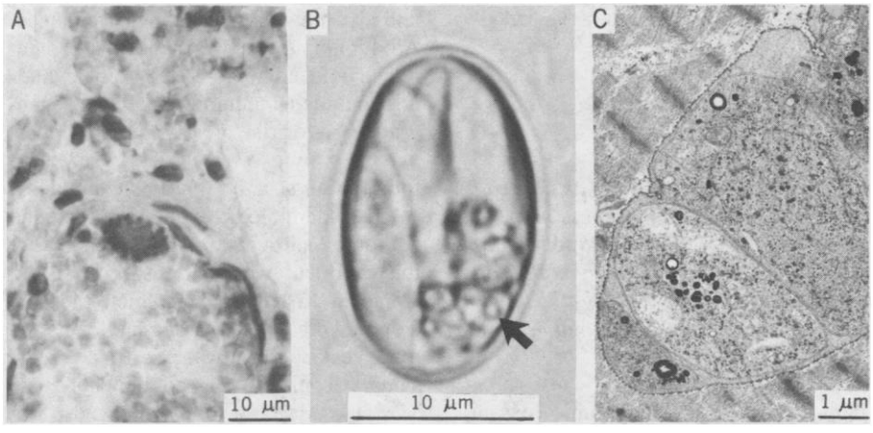

Fig. 1. (A) Photomicrograph of a histologic section of lung from a heifer with acute sarcocystosis. A schizont is present in an endothelial cell of a small pulmonary vessel. (B) Sporulated sporocyst showing four sporozoites and the granular residual body (arrow). Sporocysts were found in the dog's feces by using Sheather's sugar flotation technique. (C) Electron micrograph of a young cyst in a striated muscle cell from the diaphragm of a calf with subacute sarcocystosis. There is minimal degeneration of muscle fibrils surrounding the cyst.

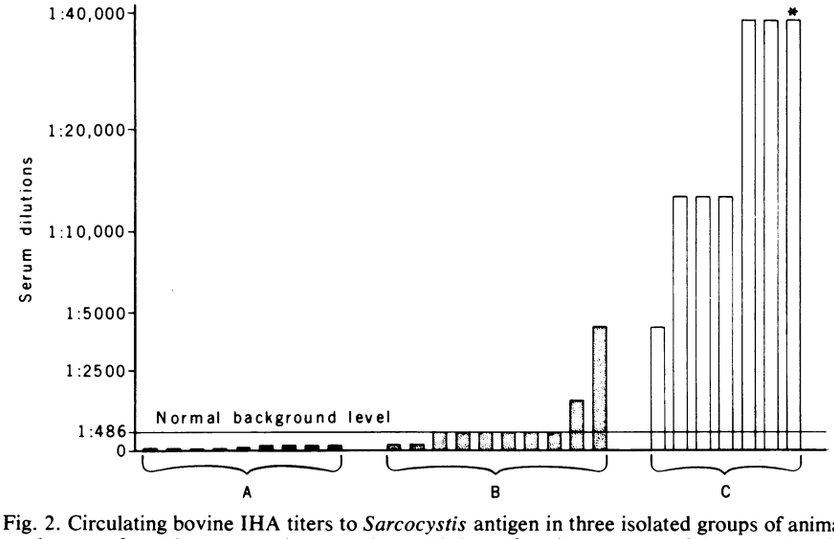

Fig. 2. Circulating bovine IHA titers to *Sarcocystis* antigen in three isolated groups of animals on the same farm. Serum samples were drawn 30 days after the necropsy of the second heifer. This heifer had a titer of 1:39,000 at the time of death (open column noted by asterisk). The remaining open columns represent six affected heifers with markedly elevated titers ranging from 1:4374 to 1:39,000. Shaded columns represent titers from ten milking cows, eight with titers ranging from 1:162 to 1:486. Two cows have elevated titers of 1:1458 and 1:4374, respectively. Solid columns represent nine 3-month-old calves with titers ranging from 1:<18 to 1:162.

P. FRELIER
Department of Pathology, New York State College of Veterinary Medicine, Cornell University, Ithaca 14853
I. G. MAYHEW
Department of Large Animal Medicine, Obstetrics and Surgery, New York State College of Veterinary Medicine
R. FAYER
Animal Parasitology Institute, Animal Research Service, U.S. Department of Agriculture, Beltsville, Maryland 20705
M. N. LUNDE
Laboratory of Parasitic Diseases, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, Maryland 20014

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