[Jpn. J. Parasitol., Vol. 44, No. 5, 391–395, October, 1995]

Sarcocystis capracanis and *S. hircicanis* from Goats in Japan

MORIHIRO SAITO[1], YUTAKA SHIBATA[1] AND HIROSHI ITAGAKI[2]

[1]*Kumagaya Meat Inspection Center, Saitama Prefecture, 179-1 Shimomasuda, Saitama 360, Japan.*
[2]*Department of Parasitology, Azabu University School of Veterinary Medicine, 1-17-71, Fuchinobe, Sagamihara, Kanagawa 229, Japan.*

(Accepted November 10, 1995)

Abstract

The heart and diaphragm were obtained from 21 adult goats slaughtered in Saitama Prefecture, Japan. Ten of the 21 goats, 48%, were positive for both infections with *Sarcocystis capracanis* and *S. hircicanis*. Cysts of *S. capracanis* (type 1) were 450–562×68–78 μm in size and had the thick wall, 2 to 3 μm thick, provided with radial striated and palisade-like villar protrusions. Two 6-month-old dogs fed with goat meat infected with *S. capracanis* cysts (type 1) shed sporulated sporocysts, 14–15×9–11 μm in size, in the feces 9 days after ingestion. *S. hircicanis* cysts (type 2) were 1,200–1,400×140–166 μm in size and had the thin wall, less than 1 μm, with hair-like protrusions on the surface. Two 6-month-old dogs fed with *S. hircicanis*-infected goat meat excreted sporulated sporocysts, 15–17×10–11 μm in size, in the feces 9 days or later after ingestion.

Key words: dog; goat; morphology; *Sarcocystis capracanis*; *Sarcocystis hircicanis*.

Introduction

Three *Sarcocystis* species utilizing goats as an intermediate host have been reported: *S. capracanis*, *S. hircicanis*, and *S. moulei*. The final host of the first two species is the dog, whereas that of the last species is the cat (Dubey *et al.*, 1989). The life cycle and pathogenicity to goats of *S. capracanis* and *S. hircicanis* are experimentally established (Collins and Charleston, 1979; Dubey *et al.*, 1981; Heydorn and Haralambidis, 1982; Pethkar and Shah, 1982; Heydorn and Unterholzner, 1983; Dubey *et al.*, 1989), however, in Japan no report has been published even on the incidence of *Sarcocystis* of goats.

We detected two species of *Sarcocystis* from slaughter goats and identified the species by morphological examination of cysts and experimental infection of dogs with cysts.

Correspondence: Morihiro Saito
斉藤守弘[1]，柴田　穣[1]，板垣　博[2]（[1]埼玉県熊谷食肉衛生検査センター，[2]麻布大学獣医学部寄生虫学教室）

Materials and Methods

Materials

Cardiac and diaphragm muscles, 50 g each, were collected from 21 goats which were slaughtered at an abattoir located in the northern part of Saitama Prefecture from March to August in 1995.

Detection of sarcocysts

Muscle blocks, 1×2×0.5 cm in size, were cut out at a right angle to muscular fibers and cysts were directly removed from the blocks in a petri dish under a microscope (Saito *et al.*, 1984). In addition to this direct method, the routine histopathological method was used for detection of sarcocysts in the muscle blocks.

Morphological observation of cysts

Fifty fresh cysts just removed from the muscles were measured with a micrometer and examined for structure of the cyst wall. A part of the cysts was fixed with 10% formalin and postfixed with 1% osmic acid solution. After fixation of the cysts were dehydrated in a series of ethanol and dried at the critical point, and then platinum was deposited onto them. The treated specimens were observed under a scanning electron microscope (Nihon Denshi, JSM-

35C) for morphology of the villar protrusions on cyst wall. For histopathological examination muscle blocks were fixed in 10% formalin and dehydrated in an ethanol series after they were ascertained to contain cysts under a light microscope. The fixed material was then embedded in paraffin, sectioned and stained with hematoxylin and eosin.

Experimental infection of dogs with cysts

Fifty cysts of type 1 and 2 each were fed two cross-breed dogs, about 6 months old, respectively with dog food, just after removed from the muscles. Other two mixed breed dogs were used as uninoculated controls. All the dogs were examined, every day PI, for sporocysts in the feces by the flotation method with saturated NaCl solution. Fifty sporocysts excreted by each dog were examined morphologically and measured by a micrometer.

Results

Ten of 21 goats examined (48%) were positive for cysts. Two types of cysts were detected from both cardiac and diaphragm muscles of all the positive animals.

Fresh cysts of type 1 were 450–562×68–78 μm in size and had the thick radial striated wall, 2–3 μm, with palisade-like villar protrusions on the surface. The same structural findings were also observed by histopathological examination. Scanning electron microscopy revealed that the palisade-like protrusions were 3–4×0.5–1 μm in size.

Fresh cysts of type 2 were 1,200–1,400×140–166 μm in size and their wall was thin and had hair-like villar protrusions on the surface. Histopathological observation showed that the wall was thin and structureless. By scanning electron microscopy,

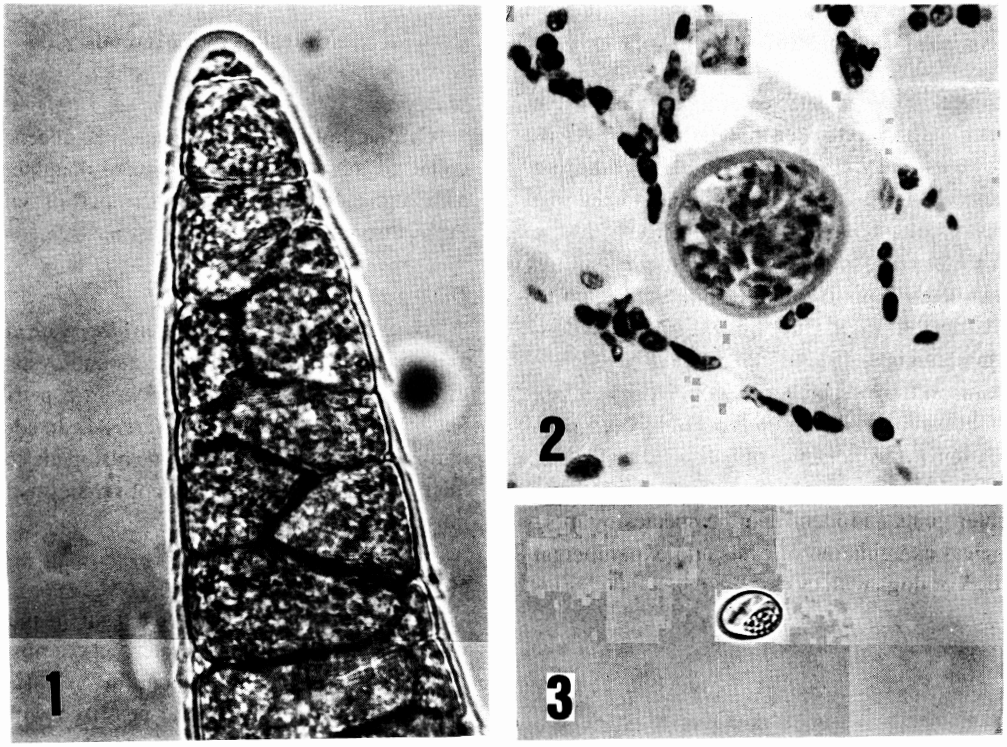


Fig. 1 A fresh *Sarcocystis capracanis* cyst (type 1) recovered from skeletal muscle of a goat. ×200
Fig. 2 A thick-walled cyst of *S. capracanis* (type 2) in skeletal muscle of a goat. H & E stain. ×400
Fig. 3 A sporocyst of *S. capracanis* (type 1 species) excreted in feces of dog. ×400

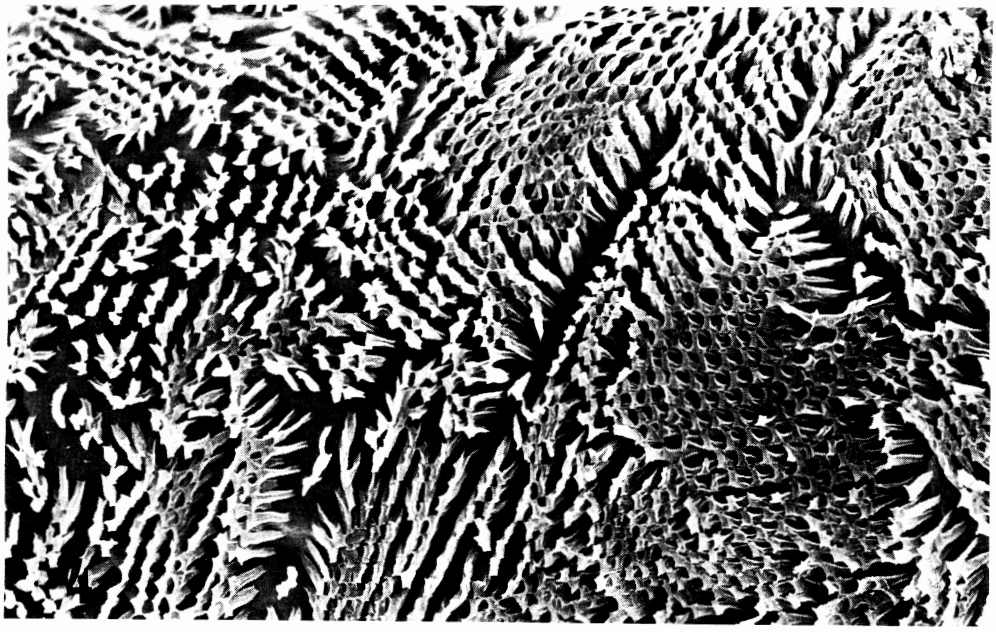


Fig. 4 Scanning electron micrograph (SEM) of *S. capracanis* cysts (type 1). Note palisade-like villar protrusions on cyst wall. ×2,000

hair-like villar protrusions, 4–5×0.2–0.3 μm in size, were observed on the surface of cyst wall. Both types of cyst contained many bradyzoites and a small number of metrocytes.

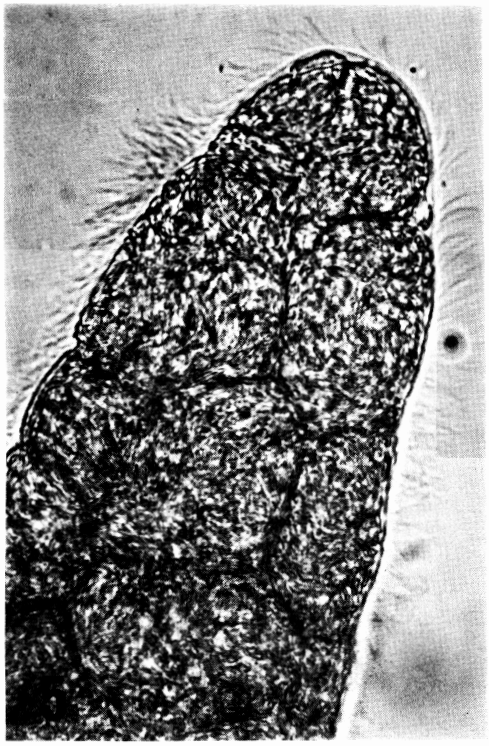
Histopathological observation revealed that no inflammatory reactions in the tissue surrounded cysts.

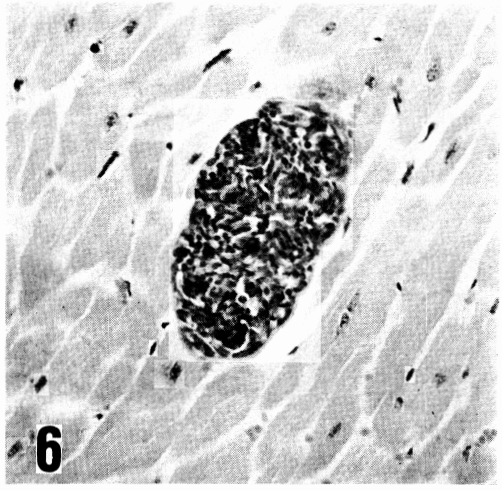
The dog fed with type 1 cysts started to shed sporulated sporocysts in the feces at 9 days PI and sporocysts were continuously passed through the experimental period of 60 days. Sporocysts were ellipsoidal, 14–15×9–11 μm in size, and contained 4 sporozoites and one large internal residual body but no Stieda body. The animal fed with type 2 cysts began to pass sporocysts in the feces at 9 days PI and continuously shed through the experimental period of 60 days. Sporocysts were 15–17×10–11 μm in size and ellipsoidal, and contained 4 sporozoites and one large internal residual body, but no Stieda body.

Discussion

In India *Sarcosystis* was detected in 432 of 650 goats examined, 66.4%, and 74.6% of the adult goats were positive for the parasite (Pethkar and Shah, 1982). The present results show that the infection rate in Japan was 48% as high as in India although the animals examined were small in number. This infection rate is lower than that in cattle but higher than those in pigs and horses in Japan (Saito *et al.*, 1984; Saito *et al.*, 1986; Saito *et al.*, 1995).

There have been known three species of *Sarcocystis* which utilize goats as the intermediate host: *S. capracanis*, *S. hircicanis*, and *S. moulei*. Two types of cysts were detected in the present survey. Type 1 cysts were almost the same as of *S. capracanis* which are more than 1,000 μm and 100 μm in length and width respectively and have the thick, more than 3 μm, wall of pallisade-like structure, provided with finger-like villar protrusions by transmission electron microscopy (Dubey *et al.*, 1989). Sporocysts in the present survey, however, were slightly smaller in length than that of *S. capracanis* but this will be caused by immaturity of the sporocysts measured because sporocysts become larger with the time passed after start of shedding. *Sarcocystis* species





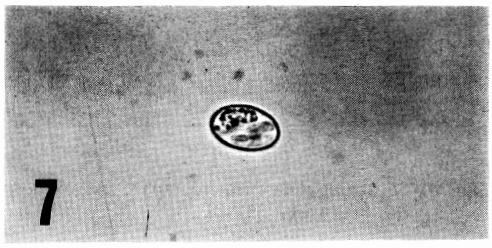


Fig. 5 Fresh *S. hircicanis* cyst (type 2) recovered from skeletal muscle of a goat. ×200
Fig. 6 A thin-walled cyst of *S. hircicanis* (type 2 species) excreted in skeletal muscle of a goat. H&E stain. ×400
Fig. 7 A sporocyst of *S. hircicanis* (type 2 species) excreted in feces of dog. ×400

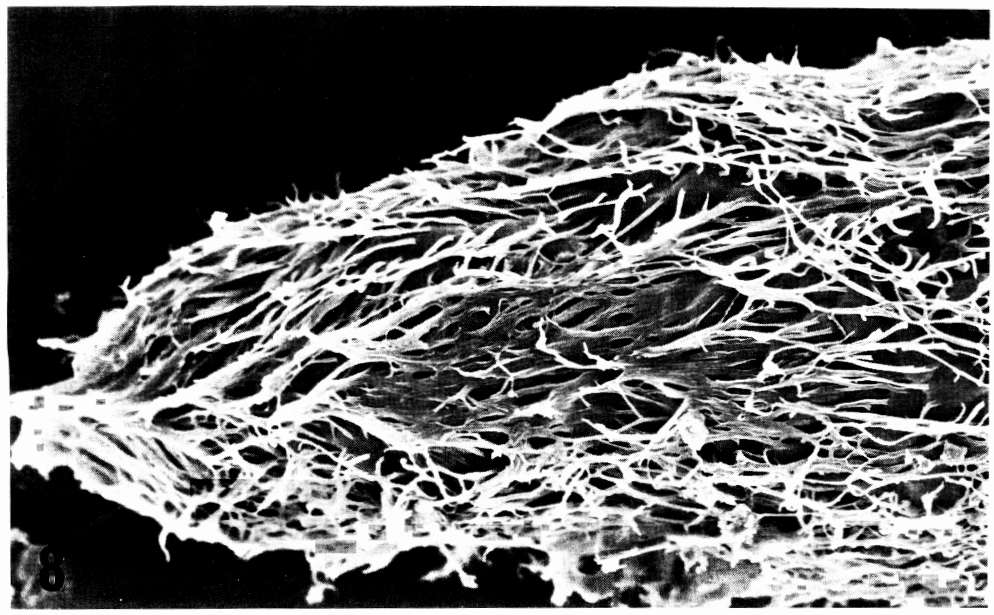


Fig. 8 Scanning electron micrograph (SEM) of *S. hircicanis* cysts (type 2). Note hair-like villar protrusions on cyst wall. ×1,800

with type 2 cyst in the present study was morphologically identical with *S. hircicanis* which has cysts, 300–2,500×20–45 μm in size, with the thin wall, provided with hair-like villar protrusions and sporocysts with a size of 15.0–17.3×10.5–11.3 μm (Heydorn and Unterholzner, 1983; Ito, 1984; Dubey *et al.*, 1989).

By the morphological characteristics of cysts, sporocysts and the final host species, type 1 and 2 *Sarcocystis* species in the present survey were identified as *S. capracanis* and *S. hircicanis*, respectively.

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