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Light Microscopic Studies of *Sarcocystis* spp. Infection in Cattle Slaughtered in Three Different Abattoirs in Metro Manila

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ABSTRACT

Examination of muscle tissues of carcasses of 370 imported Brahman breed cattle slaughtered in three different abattoirs in Metro Manila revealed an infection rate of 10.8%. Three morphologically different microscopic sarcocysts were detected; the spherical and radially striated or hirsute cysts with thick cyst wall (Type 1); the spherical to oval cysts exhibiting thinner cyst wall (Type 2) compared to Type 1; and the spindle-shaped to elongate cysts with prominent compartmentalized arrangement of zoites separated by septae. Sarcocysts morphology and their host location suggest *Sarcocystis hominis* and *Sarcocystis cruzi* as the most likely etiologic species. The infections noted may either be local or imported in origin. In the absence of any documented studies on local or imported bovine sarcocystosis in the country to date, these initial findings are valuable. However, for future studies on ultrastructural analysis of the sarcocysts and the cyst wall to confirm species identification, and experimental exposure studies to determine the probable definitive host(s) of *Sarcocystis* species are necessary.

INTRODUCTION

Earlier published reports in the Philippines have documented the presence of sarcocysts in muscles of water buffaloes typified by the Philippine water buffalo (Arambulo et al. 1972; Manuel et al. 1983) with *Sarcocystis fusiformis* as

the common etiologic species (Tongson and Molina 1978; Manuel et al. 1983; Parairo et al. 1988). In ground meat sold in public markets and supermarkets, zoites were isolated (Tongson 1960; Tongson and Pelagio 1978) which led them to suspect that it was a mixture of meat from carabaos and cattle (=carabeef). While sarcocystosis is commonly reported infecting cattle worldwide including those in New Zealand, namely *Sarcocystis cruzi, Sarcocystis hominis* and *Sarcocystis hirsuta* with canids, primates, and felids as definitive hosts, respectively (Dubey et al. 1989), in the Philippines, however, studies on bovine sarcocystosis are altogether wanting. The present study sought to find out the occurrence of *Sarcocystis* infection in cattle slaughtered in abattoirs in Metro Manila which serve as one of the main sources of meat by Filipinos.

MATERIALS AND METHODS

Muscle tissues from the esophagus, cervix, heart and lower and upper limbs of 370 cattle carcasses, age 3-11 years old were obtained from the Vitas Inspection Board, Pasay Abattoir and San Juan Municipality Slaughter House, all located in Metro Manila. Slaughtered cattle were all Brahman breed imported from Australia and New Zealand which were kept in farms and other maintenance facilities in Batangas and Pangasinan in Luzon, and Davao in Mindanao for approximately 3-4 months to fatten prior to their shipment to Metro Manila for slaughter, inspection, and distribution to various public markets, supermarkets and restaurants, as well as to meat processing companies in the Metropolitan area. Sarcocysts in muscles were determined by gross inspection and examination of unstained muscle smear preparations using Nikon photomicroscopic Model eclipse E400. Muscle tissue smears that were positive for sarcocysts were fixed in ice-cold 2.5% glutaraldehyde and were processed for TEM (Model JEOL 1010, Japan) at the Biomedical Research Unit, St. Luke's Medical Center, Quezon City.

RESULTS AND DISCUSSION

Of the total 370 cattle carcasses surveyed, 40 (10.8%) were infected with sarcocysts occurring in the muscles of the esophagus, cervix, limbs and heart. Infection rates were 19.24%, 10.40% and 2.50% among cattle surveyed at the Pasay Abattoir, San Juan Municipality Slaughter House and Vitas Inspection Board, respectively. Despite a significantly fewer female cattle used in the survey, male and female individuals showed no significant difference in their infection rates. With the generally low grade infection in cattle carcasses surveyed at the Vitas Inspection Board, some positive cases may have been missed. Also, the low infection rates in cattle noted in three different slaughter facilities may suggest either the absence of infection or existence of an early stage of infection in

imported cattle prior to shipment to the Philippines, or recent exposure to infection while the cattle were kept in the farms prior to their transport to Metro Manila.

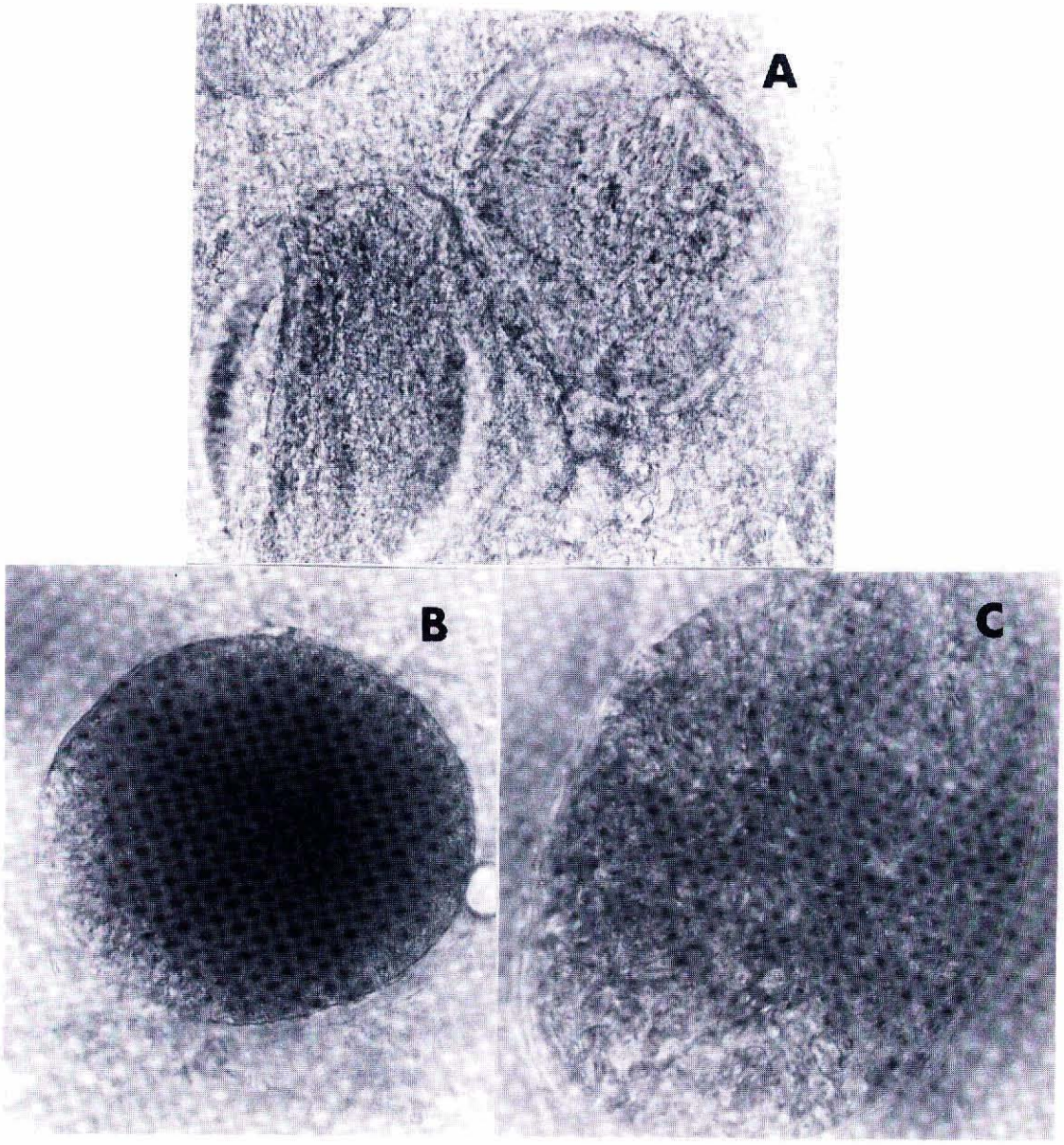

Fig. 1. Microscopic cysts from limb muscles. A: Type 1 with thick cyst wall (diameter=140 μm); B & C: Type 2 with thin cyst wall (B: diameter=45μm; C: diameter=130 μm$\times$70-80 μm). Compartmentalized arrangement of zoites is not evident.

Three morphologically different microscopic sarcocysts were observed in unstained muscle smears. Type 1 exhibits spherical and radially striated or hirsute thick-walled sarcocysts measuring about 120-145 (Fig. 1A). Type 2 is spherical to oval in shape with thin cyst wall (Figs. 1B & 1C). Type 3 exhibits either spindle-shaped or fusiform to elongate cysts with prominent compartmentalized arrangement of zoites. The spindle-shaped cysts in the striated and heart muscle tissue measure 150-200 μm by 12-24 μm in diameter (Fig. 2A); and the elongate cysts in striated muscles measure 330-450 μm by 30-40 μm (Fig. 2B). Developing or young cysts were also present.

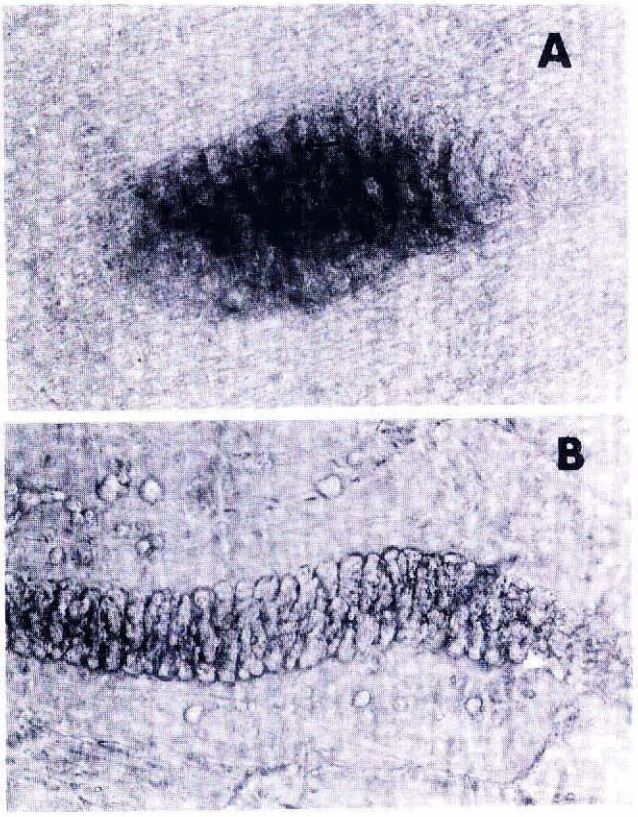

Fig. 2. Microscopic cysts from the heart (A) and limb (B) muscles. Note: compartmentalized arrangement of zoites with septal partitions (arrows). A: 150μm$\times$ 20μm; B: 410μm$\times$30μm.

Several attempts to localize sarcocysts in unstained muscle smears for histological processing and TEM were unsuccessful. The low grade infection generally noted in the muscle tissues and the possibility of damage to the sarcocysts by physical applications such as heat (Saleque et al. 1990) and pressure may have contributed to the difficulty in recovering sarcocysts. Majority of the cattle carcasses sunfeyed were in the age range 3-6 years old, and if assuming they were infected, the sarcocysts may still be developing or young and were then highly sensitive to damage. Muscle tissue smears positive for sarcocysts that were processed for TEM yielded damaged ultrathin sections and could not be used then

for light and transmission electron microscopy. Consequently, ultrastructural studies of sarcocysts cyst wall vital in the identification of the cyst type and the species (Dubey et al. 1989) was not possible.

The light microscopic description of the three sarcocysts and their host location point to *Sarcocystis hominis* (Railliet and Lucet 1891) Dubey, 1976 for sarcocysts Types 1 & 2, and *Sarcocystis cruzi* (Hasselmann 1926) Wenyon, 1926 for sarcocysts Type 3, as the most likely etiologic species. *Sarcocystis cruzi* being highly pathogenic and the most prevalent species infecting cattle worldwide including those in New Zealand, has been reported occurring in striated and heart muscle and Purkenji cells of the brain in cattle (Dubey et al. 1989). Considering that the cattle surveyed in the present study were imported from Australia and New Zealand, it is likely that they were already infected prior to their transport to the country. Considering the existence of bubaline sarcocystosis in the country, some species of which may be infective to cattle, the imported cattle may have been exposed to infection while in open farms for 3-4 months prior to their shipment to slaughter facilities in Metro Manila. In the absence of any documented studies on local or imported bovine sarcocystosis in the country to date, these initial findings are valuable. However, for future studies ultrastructural analysis of the sarcocysts and the cyst wall to confirm species identification, and experimental exposure studies to determine the probable definitive host(s) of *Sarcocystis* species documented in this paper are essential.

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