RESEARCH NOTES

SARCOCYSTIS SP. IN THE MYOCARDIUM OF A WATER BUFFALO FROM SULAWESI (CELEBES), INDONESIA

During schistosomiasis surveillance studies in Central Sulawesi tissues of an Asian water buffalo, *Bubalus bubalis*, from Puroo, Lake Lindu, Sulawesi (latitude 1° 22′ S, longitude 120° 07′ E, elevation 960 meters) were collected and preserved in 10% formalin, embedded in paraffin, sectioned at 5 microns, stained with hematoxylin and eosin, and examined microscopically. Several myocardial cysts were seen; one was in subendocardial tissue and the remaining were deeply embedded in the myocardium. Cysts generally were spindle shaped in longitudinal section (Fig. 1) and spores were seen easily. There was no evident tissue reaction around intact cysts. Neither spores nor cysts of *Sarcocystis* were found in other organs; however, neither skeletal muscle and diaphragm were examined.

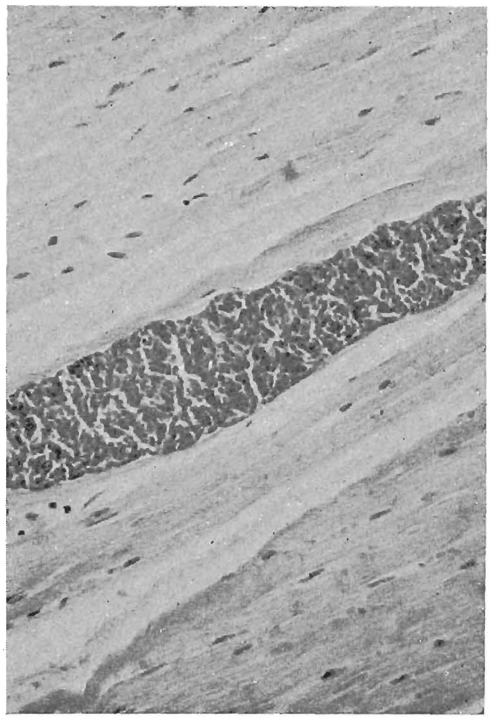


Fig. 1—Sarcosporidial cyst in myocardium. H&E. x 400.

Myocardial *Sarcocystis* is occasionally seen in bovines (Smith and Jones, 1963. *Veterinary Pathology*, 2nd ed. p. 478-481). In the Indonesian Archipelago *Sarcocystis* has been reported from water buffalo throughout the island of Java. Organisms were found in esophageal and throat tissue, skeletal muscle and in the heart (Holz, 1957. *Hemera Zoa*, *64* : 451). Recently, *Sarcocystis* was reported from rats of Central Sulawesi (Brown *et al.*, 1974. *Southeast Asian J. Trop. Med. Pub. Hlth.*, *5* : 451). The present report appears to be the first confirmation of *Sarcocystis* in bovines from Sulawesi.

Sarcocystis is occasionally reported from humans as an incidental autopsy finding. In Indonesia, Bonne and Soewandi (1929. *Geneesk. Tijdschr. v. Nederl. Indie.*, *69* : 1104) described *Sarcocystis* in a cavernous hemangioma from the lip of a Malay man, but considered the case to be without pathological significance. However, clinical manifestations associated with human infections in India have been described (McGill and Goodbody, 1957. *Brit. Med. J.*, *2* : 333).

The opinions and assertions herein are not to be construed as official or as representing the views of the Indonesian Ministry of Health or the U.S. Navy Department.

R.J. Brown. NAMRU-2, Jakarta Detachment, APO San Francisco, 96356; W.P. Carney, Naval Medical Research Institute, Bethesda, MD 20014; Moh. Sudomo, National Institute of Health Research, Jakarta; and G. Simandjuntak, Communicable Disease Control, Ministry of Health, Jakarta.

DEVELOPMENT OF FEMALE *GNATHOSTOMA SPINIGERUM* TO MORPHOLOGICAL MATURITY IN EXPERIMENTAL MICE†

There have been two reports of *Gnathostoma spinigerum* developing beyond the advanced third larval stage in white rats *(Rattus norvegicus* var. *albinus)*; the first was found by Kikuchi 1956 (quoted by Miyazaki, 1956. *Progress of Med. Parasit.* Japan, *111* : 544) and the second by Hirakawa, (1959. *Igaku Kenku, Acta Med., 29* : 895) who reported finding a single adult male *G. spinigerum* in the lung of one of more than 150 laboratory rats each experimentally infected by oral administration of 5-35 advanced third stage larvae obtained from a fresh water fish *(Ophicephalus argus)*. In Thailand, Daengsvang *et al.*, (1966. *Amer. J. Trop. Med. & Hyg., 15* : 727) have observed natural infection with *G. spinigerum* in a number of mammals, including the black rat *(Rattus rattus)*; the bandicoot rat *(Bandicota indica)*; the mongoose *(Herpetes javanicus)*; and the tree-shrew *(Tupaia glis)*. In no case was development beyond the advanced third larval stage observed.

Daengsvang, (1968. *Ann. Trop. Med. Parasit., 62* : 88) have successfully infected laboratory rodents by feeding fully developed larvae in cyclops, or by feeding advanced third stage larvae from other infected vertebrates. To date a total of 10,000 ICR mice have been infected with more than 40,000 larvae as described in that report. After penetration of the gastric and intestinal mucosa, the larvae may migrate to and encyst in visceral organs including the liver, spleen, pancreas, kidneys, adrenal glands, diaphragm and lungs. The majority, however, encyst as advanced third stage larvae in muscles and subcutaneous tissues and some in the liver and remain there almost unchanged in size and form. Some living, encysted larvae have been observed in mice at necropsy as long as 3 years after the feeding of third stage larvae. These studies have indicated the potential of mice and other rodents to act as second intermediate and paratenic hosts for *C. spinigerum*.

The finding of two mature female *G. spinigerum* in two of the aforementioned 10,000 mice is the subject of the current report. One worm was located in the subcutaneous tissue of the abdominal wall at necropsy. This worm was found 320 days after infection. The second worm was recovered from the diaphragm approximately 1 year after infection. The first worm measured 16.0 × 1.1 mm and the second 9.7 × 0.6 mm. Both worms were similar morphologically. The cephalic bulb of both worms had 8 rows of cephalic hooklets (Fig. 1) and the anterior portion of the body from the neck to approximately the middle portion of the body was covered with numerous cuticular spines (Fig. 2). Each cuticular spine had 1 to 4 terminal teeth, with 3-toothed spines being most common. The female reproductive organs of both worms appeared well developed morphologically, but no eggs were observed.

This third report of development of advanced third stage larvae to maturity in a rodent previously considered to be only a second intermediate or paratenic host is important, in that the same development could occur in man. In an area such as Thailand, where human infections with *G. spinigerum* are not uncommon, this possibility needs to be considered in the clinical management of human cases.

†The opinions or assertions contained herein are the private views of the author and are not to be construed as official or as reflecting to views of the Department of the Army or the Department of Defense.

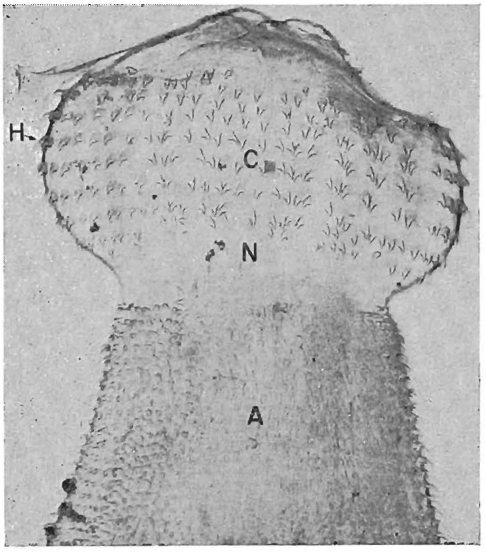


Fig. 1—Photomicrograph of the anterior end of the mature female *Gnathostoma spinigerum* from an experimental white mouse shows the cephalic bulb (C-measuring about 0.33 mm in length and 0.56 mm in width) bearing 8 rows of single-pointed cephalic hooklets (H-measuring about 13.0 × 8.0 microns), the neck (N-measuring about 0.36 mm in width) and the anterior part of the body (A-measuring about 0.5-0.9 mm in width).

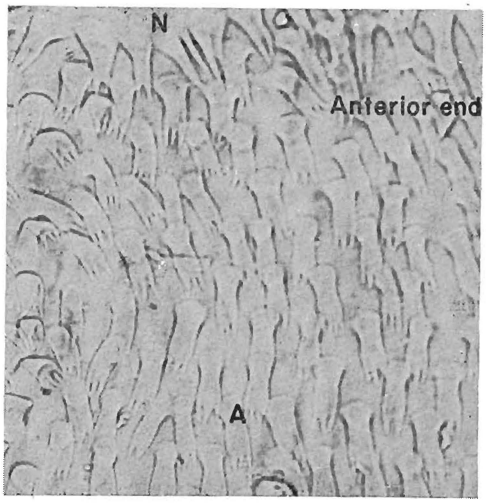


Fig. 2—Photomicrograph of the anterior body cuticular spines close to the neck of the mature female *Gnathostoma spinigerum* from an experimental white mouse shows 1 to 4 toothed spines (mostly 3-toothed spines, few 2-toothed spines are seen in the first and second rows posterior from the neck N, few 4-toothed spines are observed among the numerous 3-toothed spines).

ACKNOWLEDGEMENTS

The author expresses his appreciation to the Director, US Army Medical Component, SEATO Medical Research Laboratory for support and encouragement in this study and to Major Douglas R. Stutz, Chief, Department of Microbiology, SMRL for assistance in the preparation of the manuscript. Thanks are also extended to the author's assistants for their good services rendered to this study.

SVASTI DAENGSVANG*. Department of Microbiology, Medical Research Laboratory, SEATO Medical Project, Rajvithi Road, Bangkok 4, Thailand.

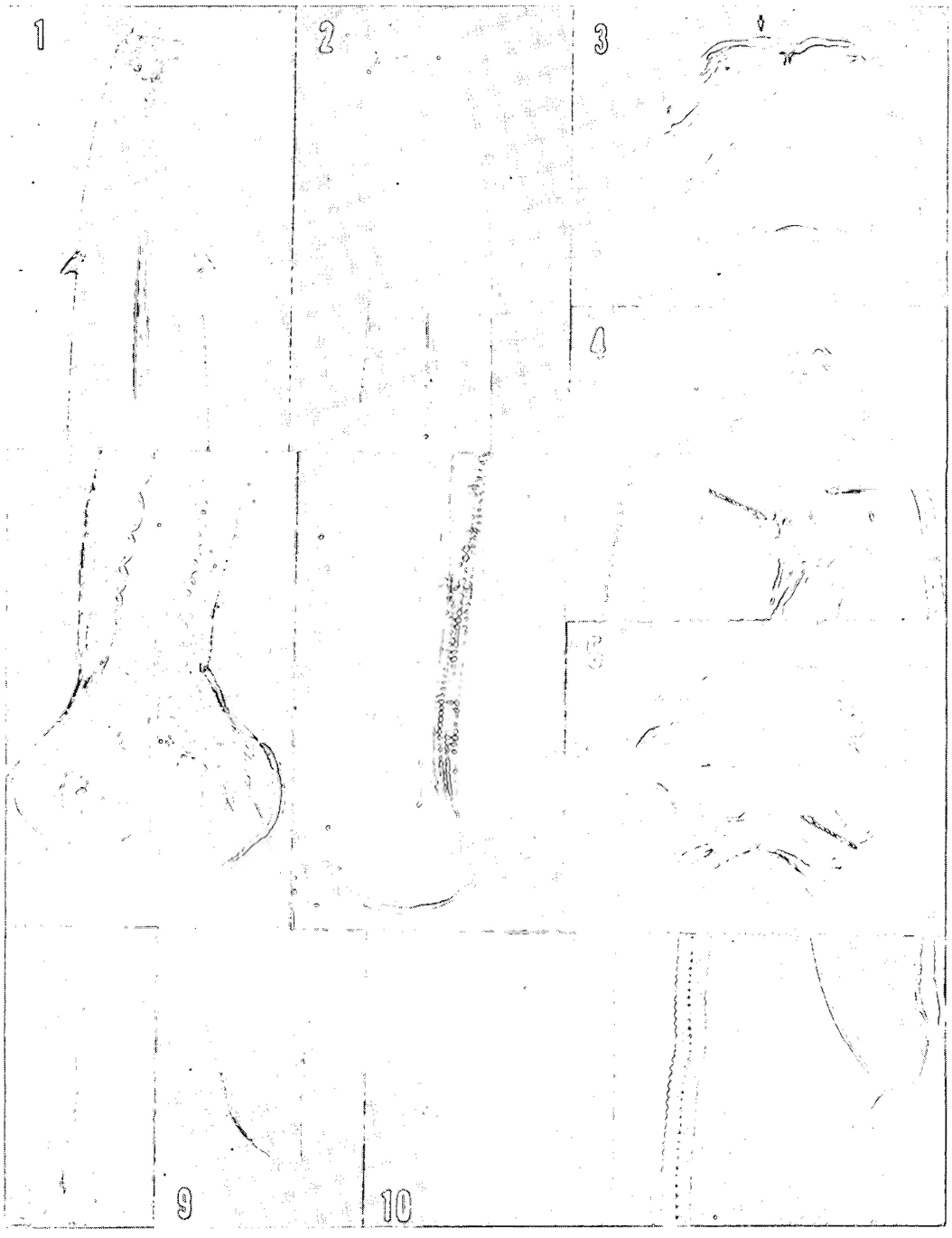
*Alternate address: SEATO Medical Project, U.S. Component, APO San Francisco 96346.

A RECORD OF *TETRAGOMPHIUS ARCTONYCIS* JANSEN, 1968 (NEMATODA: ANCYLOSTOMATIDAE) FROM THE HOG-BADGER, *ARCTONYX COLLARIS*, OF TRANG PROVINCE, SOUTHERN, THAILAND†

The genus *Tetragomphius* Baylis and Daubney, 1923 consists of parasitic nematodes characterized by the presence of a cup-shaped buccal capsule with poorly developed ventral semilunar cutting plates at the oral margin and two pairs of teeth; the mouth and buccal capsule are bent dorsally; and the male has a copulatory bursa with a pair of extremely long filiform spicules. There are 3 species in the genus: *Tetragomphius procyonis* Baylis and Daubney, 1923, from a raccoon (*Procyon* sp.) in India; *T. melis* Ohbayashi, Suzuki, and Araki, 1974, from a Japanese badger, *Meles meles anakuma*; and *T. arctonycis* described by Jansen, (1968. *J. Helminth.*, *42* : 53) who obtained more than 100 specimens from the pancreatic ducts of the mustelid hog-badger *(Arctonyx collaris)* that arrived in the Netherlands from Bangkok, Thailand. According to Jansen *(loc. cit.)*, *T. arctonycis* differs from *T. procyonis* in length of spicules, characteristics of bursa and cervical papillae, and position of vulva. The recently described *T. meles* differs from the other 2 species by having spicules intermediate in size, and smaller eggs than either of the other 2 species.

The brief summary of the present finding of adult *T. arctonycis* is as follows: A thin, adult, female hog-badger *(Arctonyx collaris)* that died in Trang Province of Southern Thailand was submitted to Major H. Rozmiarek of the Department of Veterinary Medicine, SEATO Medical Research Laboratory, Bangkok, by Mr. Klaus Berkmuller of the Nature Education Center, Forest Experiment Station, Khaochong, Trang, for a necropsy. Grossly the skin of the animal was dry and scaly and the mucous membranes were yellowish. The trachea and bronchi were filled with a foamy white exudate. There were numerous white foci about 2.0 mm in diameter disseminated over the capsular surface of the liver. The stomach was empty. The mucosal surface of the urinary bladder was diffusely reddened. Surrounding the pancreatic duct was a large firm mass, about 3 cm in diameter, with an irregular surface. When this mass was incised, numerous gritty foci were found embedded in it. In the center of the mass was the dilated pancreatic duct containing 15 white parasitic nematodes that were identified later as adult *T. arctonycis* (10 females and 5 males). Microscopic examination of the tissue surrounding the pancreatic duct revealed mature fibrous connective tissue with a few foci of mineralization. The worm specimens were later sent to the Department of Microbiology, SEATO Medical Research Laboratory, for further identification. Subsequently, some adult worms were sent to U.S. Department of Agriculture, ARS, Animal Parasitology Institute, Beltsville, Maryland, U.S.A. They were deposited in the U.S. National Parasite Collection as USNM Helminthological Collection No. 72842. The average size of three females was 19.0 × 0.4 mm (variation 18.2-20.0 × 0.3-0.5 mm) and that of three males was 17.0 × 0.3 mm (variation 13.3-20.1 × 0.3 mm). The females had numerous eggs inside the long uterus. Ten eggs averaged 81 × 47 microns (range 75-85 × 42-50). In the Jansen report *(loc. cit.)*, only one egg (98 × 49 microns) was seen in the uterus. The paired filiform spicules were about 3/4 of the length of the worm, measuring 15.1 mm long in a worm

† The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting the views of the Department of the Army and Department of Defense.



Figs. 1-11—*Tetragomphius arctonycis.* Fig. 1. Head, esophagus, and hook-like cervical papillae, ventral view, x 60. Fig. 2. Dorsally bent head, lateral view, x 60. Fig. 3. Semilunar cutting plates (arrow) of buccal capsule, ventral view, x 290. Fig. 4. Dorsally bent head showing dorsal gutter (left arrow), between dorsal teeth, and ventral teeth (right arrow), lateral view, x 290. Fig. 5. Head showing pair of dorsal teeth (arrow), ventral view, x 290. Fig. 6. Male tail showing short bursa and part of exceptionally long spicules, ventral view, x 75. Fig. 7. Male genital cone showing spicules passing through lightly sclerotized spicular canal, ventral veiw, x 250. Fig. 8. Fused spicule tips, x 105. Fig. 9. Female tail, lateral view, x 60. Fig. 10. Vulva, lateral view, x 60. Fig. 11. Cuticle, at esophago-intestinal junction, x 160.

measuring 20.1 mm; whereas, Jansen *(loc. cit.)* reported that the spicules extended from the end of the esophagus to the tip of the genital cone and measured 8 to 15.5 mm long in specimens 9.0-17.0 mm long. Figures 1 to 11 illustrate the morphologic characteristics of the nematode.

The present report is only the second of *T. arctonycis*. In both reports, the nematodes were found in the pancreatic duct of the adult hog-badger *(Arctonyx collaris)*. Rao and Acharjyo, (1972. *Current Science*, *41* : 614) reported another nematode, *Arthrostoma longespiculum* (Maplestone, 1931) Schmidt and Kuntz, 1968, in a fibromatous mass in the pancreas of a hog-badger in India. This nematode can be easily distinguished from *Tetragomphius arctonycis* on the basis of the much smaller spicules (1.2-2.2 mm long), the smaller eggs (60-66 mm long), and the smaller body size (3-5 mm long) of *A. longespiculum*.

ACKNOWLEDGEMENTS

Acknowledgements are extended to Mr. Klaus Berkmuller and Major H. Rozmiarek for their agreements to publishing the information appearing in this paper. Acknowledgements are also extended to Mr. Paisarl Yingyourd, Department of Microbiology, for technical assistance; to Capt. Garrett S. Dill, Jr. VC, Department of Veterinary Medicine, for the information concerning microscopic changes of the tissue; and to the Director and Chiefs of Departments of Microbiology and Veterinary Medicine, U.S. Army Medical Component, SEATO Medical Research Laboratory, for their interest and support in this joint study and report.

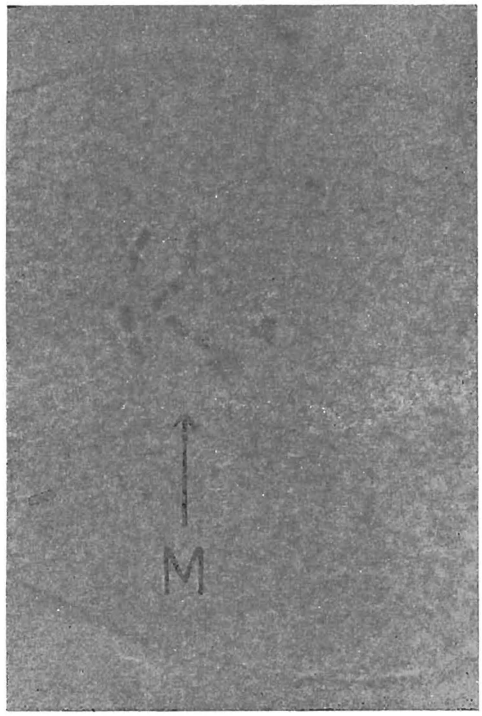
SVASTI DAENGSVANG*,+, MARKPOL TINGPALOPONG**, J. RALPH LICHTENFELS***. *Department of Microbiology, **Department of Veterinary Medicine, Research Laboratory, SEATO Medical Project, Rajvithi Road, Bangkok, Thailand. ***Animal Parasitology Institute, Agricultural Research Service, USDA, Beltsville, Maryland 20705, U.S.A.

+Alternate address: SEATO Medical Project, U.S. Component, APO San Francisco 96346.

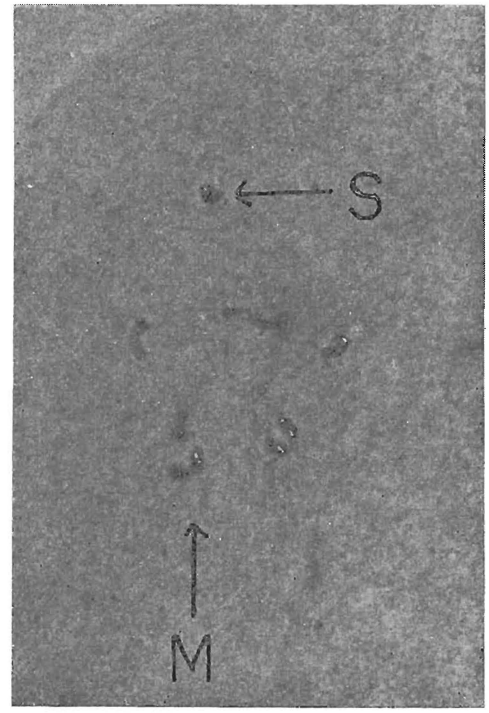
THE CHROMOSOME NUMBERS OF *ANCYLOSTOMA TUBAEFORME* ZEDER, 1800

Koidzumi *et al.*, (1926. *Trans. Far Eastern Ass. Trop. Med. 6th Congress*, *1* : 335) found that the haploid number of the chromosome seemed to be 12 in the human *Ascaris* and 13 in the pig *Ascaris*. Walton, (1959. *J. Parasit.*, *45* : 1) showed that worms which belonged to the same genus might have a different chromosome number; he also stated that all species of the order *Strongylata* with the exception of *Filaroides mustelamum* have chromosome number 2N = 12. Le Jambre and Georgi, (1970. *J. Parasit.*, *56* : 131) found that the diploid chromosome number of *Ancylostoma caninum* was 2N = 12. The chromosome number of *Ancylostoma tubaeforme* have never been reported. For this reason, the present study was undertaken. All specimens were collected from cats in Brisbane, Australia and chromosomes were stained by the technique of Le Jambre, (1968. *Trans. Amer. Microsc. Soc.*, *87* : 105). It was found that the diploid chromosome number in *A. tubaeforme* was 2N = 12. (see Figs. 1-2).

PRASERT SETASUBAN. Department of Parasitology, University of Queensland, Brisbane, Queensland, Australia.



1



2

Figs. 1-2—Photomicrograph of an egg of *A. tubaeforme* showing 12 chromosomes in metaphase II. M = Six pairs of chromosomes in metaphase II. S = Sperm nucleus.

ABNORMALITY IN SPECIMEN OF *ANCYLOSTOMA TUBAEFORME* FROM CAT IN AUSTRALIA

The form and number of the ventral teeth on the ventral margin of the mouth are important in identification of species in the genus *Ancylostoma*. The number of ventral teeth is normally constant for each species; nevertheless variation in number of ventral teeth can occur. Rep, (1963. *Trop. Geogr. Med.*, *15* : 173) found that *Ancylostoma duodenale* is characterized by two pairs of ventral teeth; however in 4.3% of his specimens each median ventral tooth had an additional "tiny tooth" on the internal edge. This did not necessarily indicate a separate species but might have been a manifestation of intraspecific variation or caused by fixation. Such a variation has been found in one of more than 1,000 specimens of *Ancylostoma tubaeforme*, all collected from the small intestines of cats in Brisbane, Australia. In the unusual specimen, the posterior ventral tooth on the right side was subdivided into two (see Fig. 1). The teeth on the left side were normal. As far as is known, this is the first report of abnormal teeth in *Ancylostoma tubaeforme*.

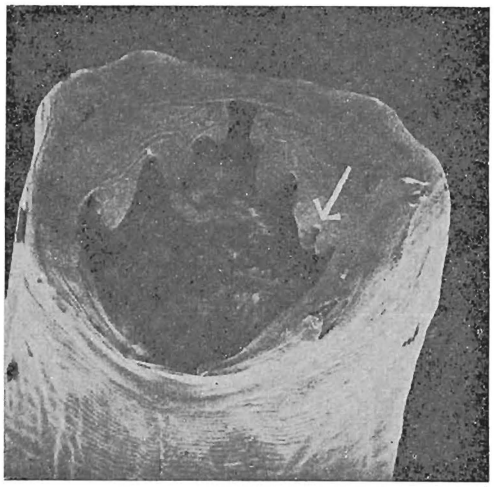


Fig. 1—Scanning electron microscopic photograph of *A. tubaeforme* showing abnormal tooth, x 540.

PRASERT SETASUBAN*. Department of Parasitology, University of Queensland, Brisbane, Queensland, Australia.

*Present address: Department of Helminthology, Faculty of Tropical Medicine, Bangkok 4, Thailand.