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The Occurrence of *Bertiella studeri* (Cestoda: Anoplocephalidae) in Japanese Macaque, *Macaca fuscata*, from Mie Prefecture, Japan

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Abstract

Three complete tapeworms and strobila were detected respectively from two Japanese macaque, *Macaca fuscata*, which were caught in Mie Prefecture, Japan. Morphological features were comparable to characteristics of *Bertiella studeri* (Blanchard, 1891). These are the first two confirmed cases of infection with the cestode in Japanese macaque in Japan.

Key words: Cestode, *Bertiella studeri*, Japanese macaque, Japan

Bertiella studeri, a cyclophyllidean tapeworm of the family Anoplocephalidae, is found in the intestine of monkeys and other primates in South Asia, Philippines, Mauritius Is. and Africa (Spasskii, 1951). The life cycle of this tapeworm was elucidated with the mite, *Scheloribates laevigatus*, being shown as an experimental intermediate host (Stunkard, 1940).

Human infection is believed to occur by accidental ingestion of mites containing cysticercoid larvae of this tapeworm and many cases have been reported mainly from tropical areas (Faust *et al.*, 1971).

Recently, a case of a 3-year-old female child living in Mie Prefecture, infected with *Bertiella* sp. (probably *B. studeri*), was reported for the first time in Japan (Kojima *et al.*, 1992). Successively, one more human case of a 2 year-old female child living in Osaka, infected with *B. studeri*, was reported (Iseki *et al.*, 1993). However, their sources of infection are unknown because both patients have never been abroad. So, in order to clarify the infectious source of the first case, we investigated the stools of Japanese macaque, *Macaca fuscata*, which were sometimes caught as vermin in the Suzuka Mountains of Mie Prefecture and brought to the Institute of Laboratory Animals, Mie University School of Medicine. We obtained three tapeworms from the intestine of a Japanese macaque and identified them as *B. studeri*. We report here with photographs the morphological characteristics of these tapeworms.

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Materials and Methods

Two out of four Japanese macaque caught from January to April 1993, periodically passed the same kind of strobila. We obtained the intestine of one monkey (12kg, male) sacrificed for other studies. The intestine was dissected and three mature undamaged cestodes were recovered. They were located in the small intestine about 92cm from the stomach. When recovered from the intestine, the worms were very active in PBS. Eggs were discharged through the uterine pore from the gravid proglottids when the musculature of the proglottids contracted. Specimen A was cut into pieces, some of which were stained with aceto-carmine and others were cut into serial transverse and horizontal sections. Specimens B and C were preserved in 10% formalin.

Description of worms

The length (cm), maximum width (mm) and number of proglottids of the specimen A are 36, 12 and 873, those of the specimen B are 37, 17 and 957 and those of the specimen C are 40, 15 and 967,

respectively (Fig. 1). However, the worms are very flexible and sometimes reached a length twice as long as the values described above. Head of the worm is unarmed. A scolex, average 647 μm in length and 730 μm in width, with 4 suckers (270 μm in diameter) is present (Fig. 3). The neck is very short. All proglottids are much wider than long; the number of proglottids per 1 cm for young strobila, mature strobila and gravid strobila are 24, 17 and 14, respectively.

The musculature of the strobila is well developed. The longitudinal muscular fibers form bundles disposed in two layers, particularly in gravid proglottids (Fig. 17). The number of fibers in a single bundle varies between 3 and 52. Two pairs of longitudinal excretory canals on each side are distinctly visible in all parts of the strobila (Fig. 10). The ventral excretory canal is much larger than the dorsal one and ventral excretory canals of both sides are connected in the posterior part of each proglottid by a wide transverse excretory canal (Fig. 13).

Each proglottid has one set of male and one set of female organs (Figs. 6–9). Each proglottid has more than 150 testes (average 89.5×52.5 μm in size) situated dorsally in the anterior part of the proglottid between the longitudinal excretory canals, forming up to 4–5 layers in the longitudinal direction (Figs. 7 and 10). The oval cirrus sac (average 586×198 μm in size) filled with spermatozoa lies anteriorly and dorsally to the vagina (Figs. 6, 9 and 10). It is a strong muscular organ with a 20 μm wall. The cirrus projects from the cirrus sac in some proglottids (Fig. 9). The vagina is 570×280 μm in size, surrounded by a thick layer of gland cells and lies posteriorly and ventrally to the cirrus sac (Figs. 6 and 12). The genital pores are irregularly alternate; often two but never more than two successive pores are observed on the same side (Fig. 14). The pore is about midway between the anterior and posterior margins of the proglottids. The ovary consists of a mass of clavate lobes which extend from the compact central part of the organ (Fig. 8). It measures 0.7 to 0.9 mm in diameter and lies about its width from the genital pore. The vitellaria is a C-shaped organ and lies beneath the ovary (Figs. 6 and 8). The uterus extends as a tube across the entire central field of the proglottid (Fig. 11) and forms pouches which occupy most of the mature proglottid (Figs. 2 and 16). Finally, the wall of the proglottid is broken on one or both sides, with the formation of a uterine pore (Fig. 17).

Fresh eggs, 43.6 μm average diameter, discharged from the uterine pore are turbid and look like soccer balls due to wrinkles on the surface (Fig. 4). However they become transparent with time and the pyriform apparatus and onchosphere clearly appear (Fig. 5). The pyriform apparatus is 25.0 to 32.5 μm long (average 30.3 μm) and 20 to 23.8 μm wide (average 22.4 μm). The onchosphere, which moves very actively, has an average diameter of 18.9 μm and has 6 slender hooks, about 7.8 μm in length.

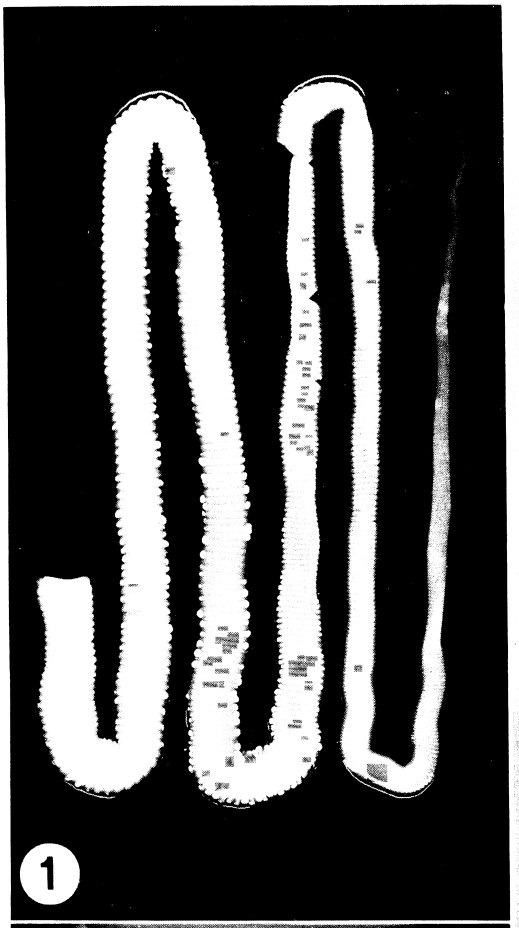
Discussion

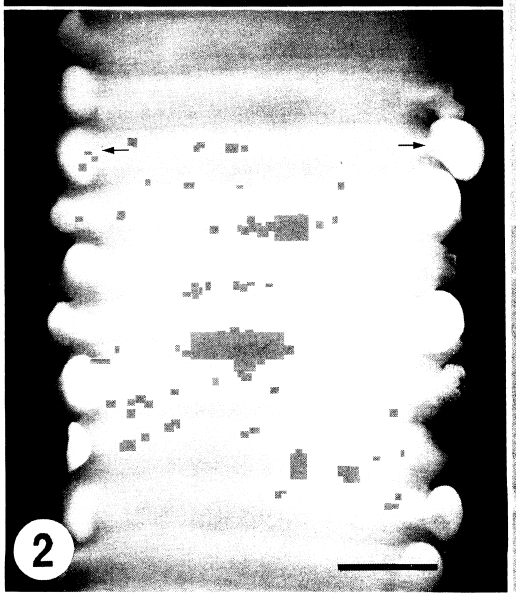
Twenty nine species of the genus *Bertiella* were reported by 1986 (Schmidt, 1986). Based on the key for determination of genera of Anoplocephalinae (Schmidt, 1986) and species of the genus *Bertiella* (Spasskii, 1951), morphological features of the present specimens resembled those of *B. studeri* or *B. mucronata*. Many aspects of the morphological differences between *B. studeri* and *B. mucronata* have been discussed (Cram, 1928; D'Alessandro *et*

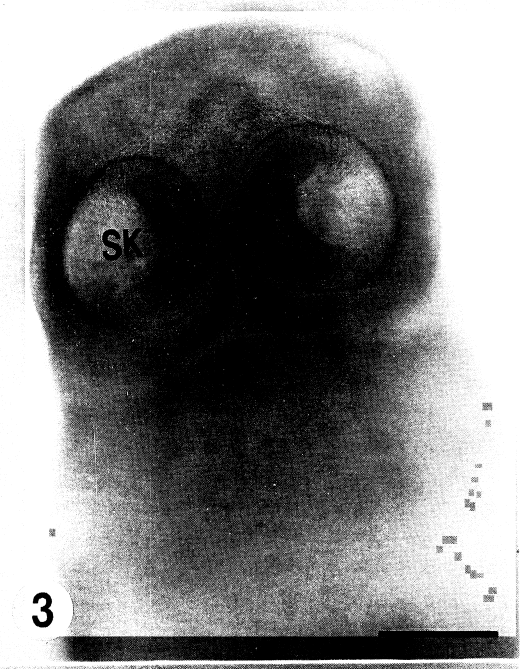
Abbreviations

CR; cirrus, CS; cirrus sac, DE; dorsal excretory canal, EP; embryophore, ES; egg shell, LM; longitudinal muscle bundles, OS; onchosphere, OV; ovary, SK; sucker, SP; spermatozoa, TE; transverse excretory canal, TM; transverse muscle, TS; testes, UT; uterus, VE; ventral excretory canal, VG; vagina, VT; vitellaria

Fig. 1 Whole specimen (A) of *Bertiella studeri* (36cm).
Fig. 2 Uterine pouches (arrow) on both sides of gravid proglottid (scale: 2mm)
Fig. 3 Enlargement of scolex (700×620 μm in size) with 4 suckers (270 μm in diameter) (scale: 200 μm).
Fig. 4 Egg from gravid proglottid (44 μm in diameter) (scale: 20 μm).
Fig. 5 Egg with pyriform apparatus (30×22 μm in size) and onchosphere (19 μm in diameter) (scale: 20 μm).



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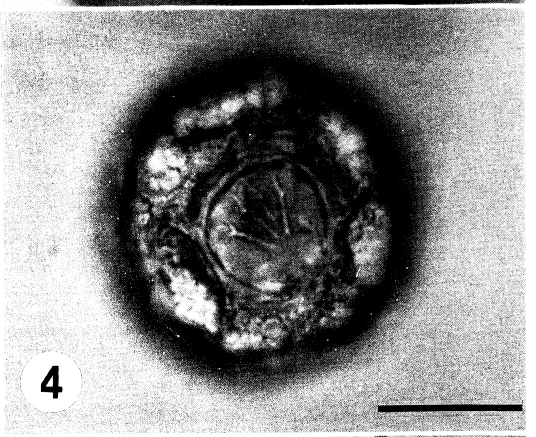

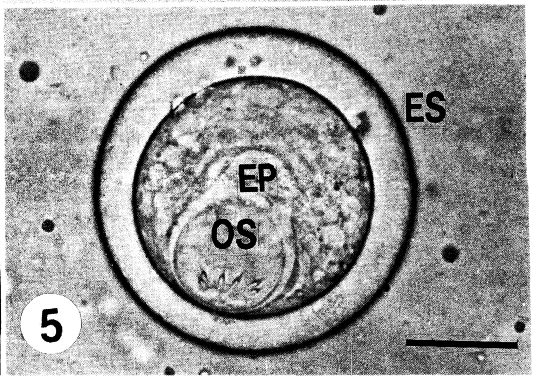
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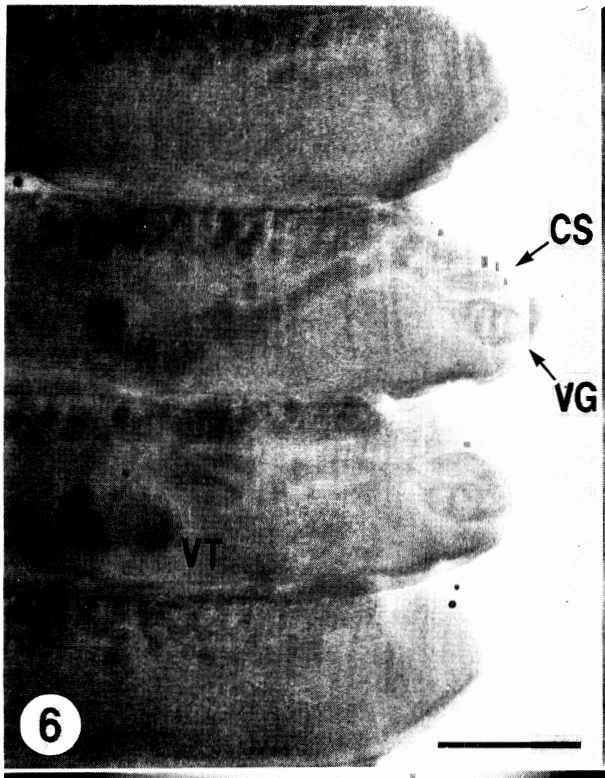
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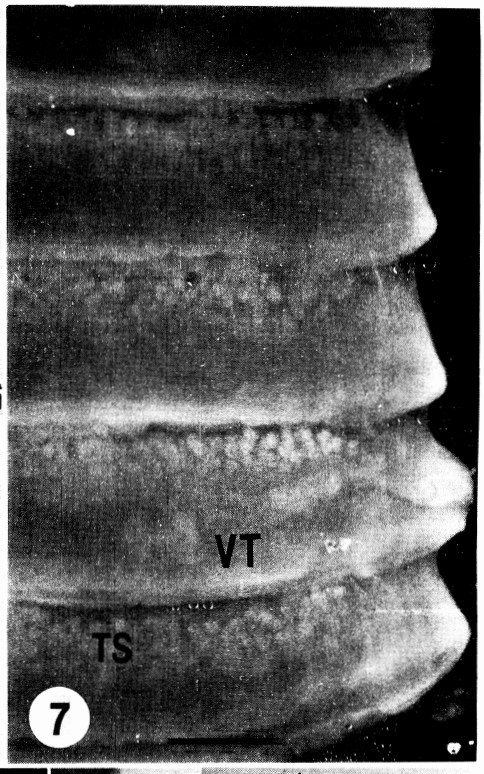
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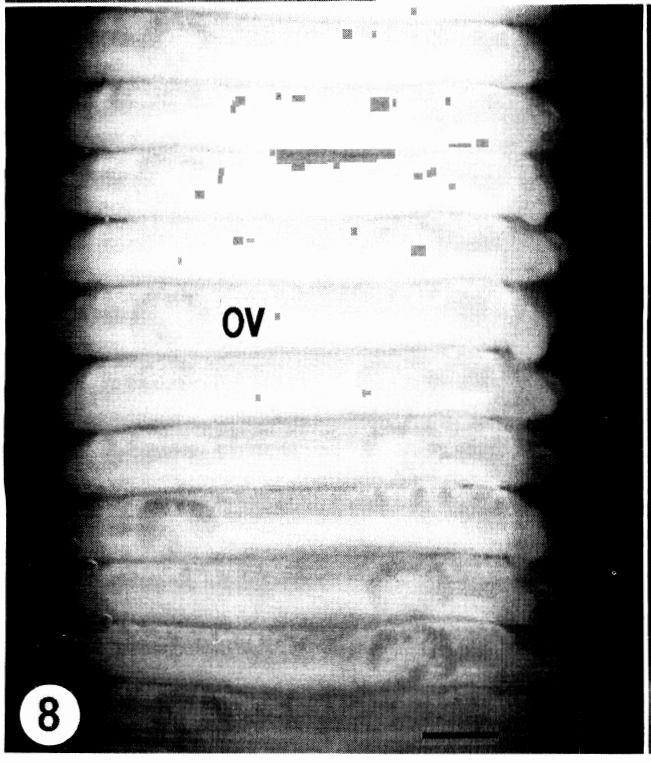
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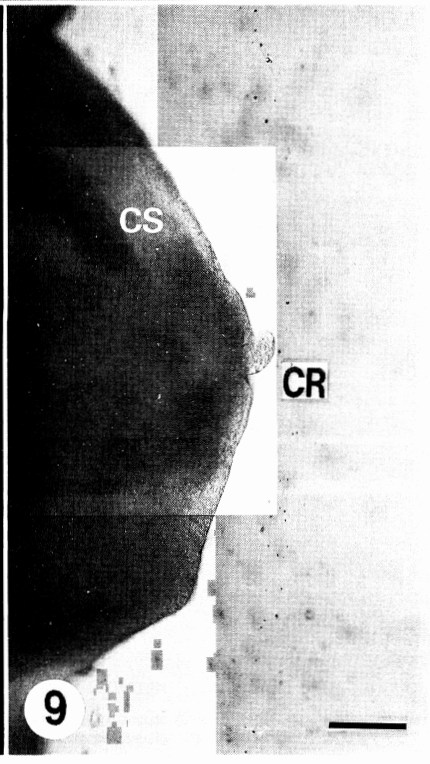
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CR
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al., 1963; Stunkard, 1940) though both species are regarded as identical by some authors (Cameron, 1929; Adams and Webb, 1933). The morphological features of the present specimens differed from those of *B. mucronata* in possessing the following; strongly developed muscular cirrus sac, weakly developed vagina and larger eggs and pyriform apparatus. Moreover, *B. studeri* has been reported only in the Eastern Hemisphere and *B. mucronata* has been found only in the Western Hemisphere. Therefore, we identified the present specimens as *B. studeri*.

In Japan, tapeworms of the genus *Bertiella* have been found in monkeys imported from tropical areas (Yoshida, 1907; Hashimoto and Honjo, 1966; Sawada and Kifune, 1974). Although eggs of *Bertiella* were also found in Japanese macaque, *M. fuscata*, adult worms, unfortunately, were not found (Tanaka *et al.*, 1962; Kagei and Hasegawa, 1974). Therefore, this is the first report which proves the occurrence of *B. studeri* in Japan. Oribatid mites, *S. laevigatus, Gulumna* sp., *Scutovertex minutus* and *Achiptleria coleptratus* are shown to be the intermediate hosts of *B. studeri* by experimental infection (Aoki, 1980). Among them, *S. laevigatus* is a strong possible intermediate host because it exists in Japan and ingested eggs grow to cysticercoid larvae in these mites. Two Japanese macaque from Suzuka Mountains in Mie Prefecture were infected with *B. studeri*. It is likely that the life cycle of this worm is completed through an intermediate host in Suzuka Mountains. This intermediate host has not yet been determined.

Among 29 species of the genus *Bertiella*, only two species, *B. studeri* and *B. mucronata* are known to infect man (D'Alessandro *et al.*, 1963). Since the first human case was reported by Blanchard (1913) about 51 human cases (Kagei, personal communication), including two children from Japan described above, have been reported. The number of reported human cases from Indonesia is the highest, being 12 cases (Kagei *et al.*, 1992). The first patient in Japan lives in a small town located at the foot of Suzuka Mountains, she might have been infected with *B. studeri* near there. Two human cases occurred one after another in Japan. Accordingly, tapeworms must be identified with great care as new human cases are likely to occur in the future.

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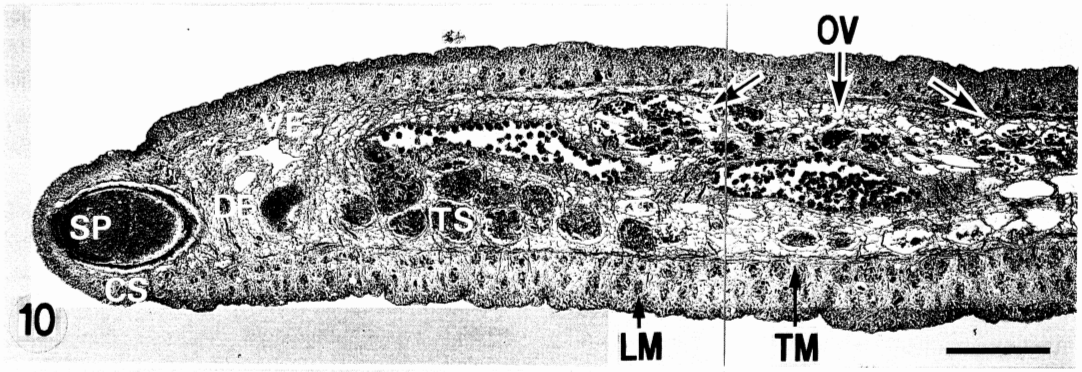
Figs. 6–8 Mature proglottid. Reproductive organs are irregularly alternate, testis is anterior, and cirrus sac and vagina are almost same size (scale: 1mm).
Fig. 9 Cirrus from mature proglottid (scale: 100 μm).



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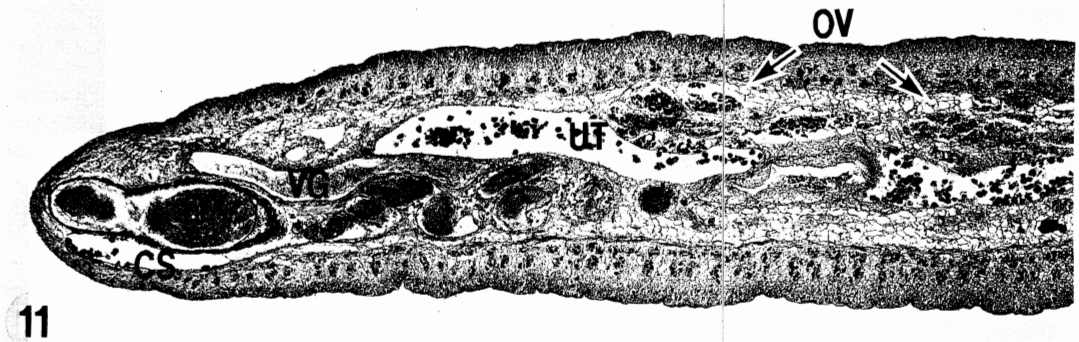

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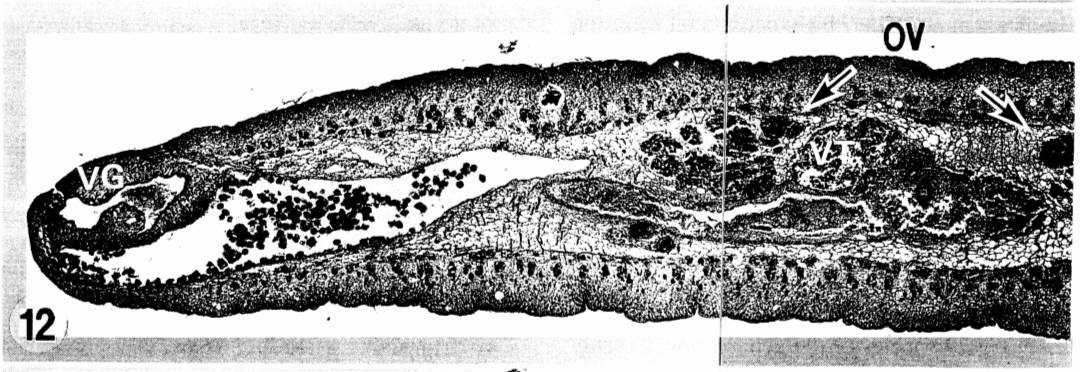

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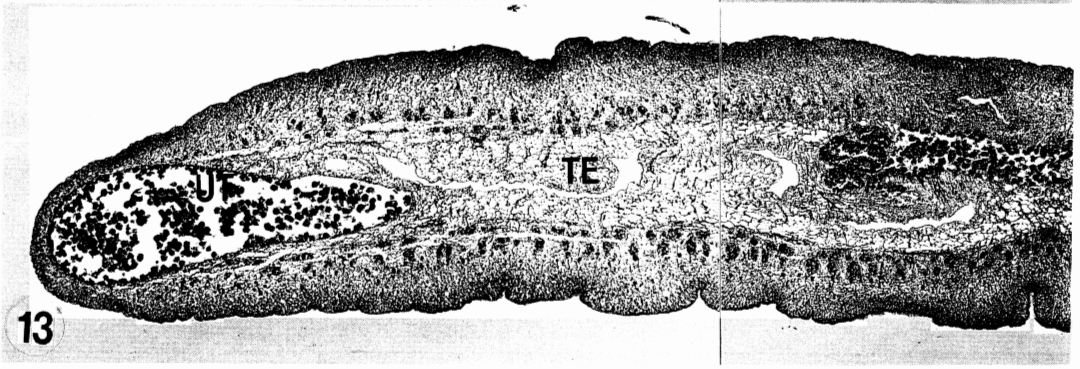

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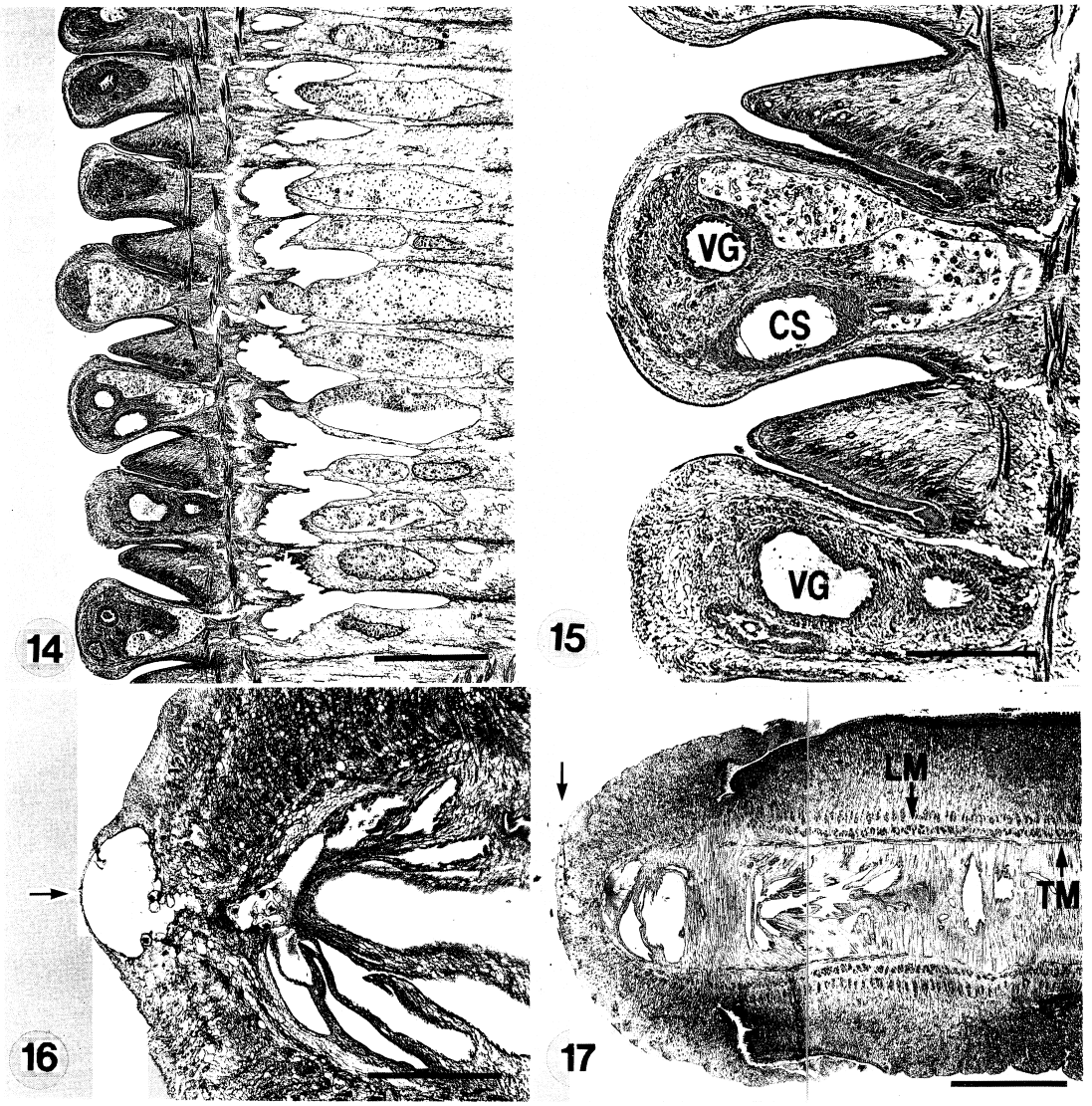

Fig. 14 Horizontal sections of mature proglottids. The proglottids with round edges are proglottids having genital pore. Proglottids with triangular edges have a genital pore on the other side (scale: 200 μm).

Figs. 16–17 Transverse section of gravid proglottid (3mm in thickness).

Fig. 16 A portion of uterine pouch (arrow) (scale: 400 μm).

Fig. 17 Uterine pore (arrow) (scale: 1mm).

Figs. 10–13 Transverse sections of mature proglottid cut from anterior to posterior (520 μm in thickness) (scale: 200 μm).

Fig. 10 Testis is dorsally.

Fig. 11 Testis almost disappears, a portion of vagina appears and ovary is separated on both sides of vitellaria (still not present).

Fig. 12 Cirrus sac disappears and vagina appears.

Fig. 13 Transverse excretory canal appears.

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