Comparison of Scanning Electron Microscopy on Baylisascaris transfuga, Toxascaris leonina and Ascaris lumbricoides (Nematoda: Ascarididae)

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Key words: SEM, Baylisascaris transfuga, Toxascaris leonina, Ascaris lumbricoides

Genus Baylisascaris was created by Sprent (1968), comprising seven species which had formerly been included in either genus Toxascaris or Ascaris of the family Ascarididae.

Prior to the creation of this genus, for at least some of the species, the distinctions between them when studied under light microscopy were the cause of considerable debate concerning their classification (Mozgovoi, 1953; Sprehn and Haakh, 1956; Hartwich, 1962).

Recently we reported the finding of *B.* transfuga in the wild black bear of Japan (Uni et al., 1981). As a supplementary study to that report, we describe herein the details of the external structures of three genotypes—*B.* transfuga, *T.* leonina and *A.* lumbricoides—examined under scanning electron microscopy (SEM) in order to obtain a better knowledge of the differences and similarities between them.

Materials and Methods

B. transfuga: One thousand fifty specimens, 104–274 mm long, collected from 2 polar bears (*Thalarctos maritimus*) by anthelmintic worming treatments at Tennoji Zoological Gardens, Osaka. Five specimens, preserved in 70% ethyl alcohol, were used.

T. leonina: Forty specimens, 62–102 mm long, collected from the small intestines of two dogs in the animal laboratory of this medical school. Ten specimens were used.

A. lumbricoides: Twelve specimens, 123–258 mm long, obtained by anthelmintic treatments given 2 patients in the hospital of this medical school.

The anterior and posterior parts of the adult worms and the eggs from the uterus were prepared for SEM examination by the same method previously described (Uni and Takada, 1975).

Results

The Anterior Part

En face view of head of B. transfuga showed three lips with dentigerous ridges, amphidial pores, internal labial papillae, lateral papillae, subventral labial papillae and dorsal labial papillae (Fig. 1). The teeth of this species were larger and blunter than those of A. lumbricoides. Those of T. leonina were much fewer in number and were arranged with greater distance between each other. Total number of teeth counted on the three lips was approximately 650 in B. transfuga, 730 in A. lumbricoides and 210 in T. leonina, respectively

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(Figs. 2, 13, 22). Lateral papillae near amphidial pores showed a walnut-like surface in all specimens of the three species (Figs. 3, 14, 23). Six internal labial papillae were recognizable as small pores on the tops of the three lips in each species. In *B. transfuga* and *T. leonina* the dorsal and subventral labial papillae exhibited distinctly divided surfaces (Figs. 4, 15), while in *A. lumbricoides* they were undivided, appearing like a single elevation (Fig. 24).

The Cervical Part

Cervical alae of *B. transfuga* and *T. leonina* were salient (Figs. 1, 12). In *A. lumbricoides* the alae, though vestigial, could be found on the lateral lines in this part in almost all specimens examined by SEM (Fig. 25). Deirids of *B. transfuga* and *T. leonina* were smaller than those of *A. lumbricoides*, all being situated on the dorsal side of the alae (Figs. 5, 16, 25).

The Posterior Part

Pericloacal area of male B. transfuga showed 4 to 5 transverse lines composed of tiny spines precloacally, and 9 to 10 such lines postcloacally (Figs. 6, 7). Such a unique configuration, having a rather rough surface, was found in all male specimens of A. lumbricoides (Fig. 27 *). (A similar structure was also found in a specimen of A. suum taken from a pig). In T. leonina this area closely anterior and posterior to cloaca appeared as a rather smooth cuticular elevation (Fig. 20 *). Regarding spicule, the surface of their distal part was wrinkled in B. transfuga (Fig. 7), but was smooth in other species. Precloacal central papilla could be recognized at the top of the area in all specimens of three species (Figs. 7, 20, 27). Postcloacally, in B. transfuga there were two pairs of double papillae, the second one smaller (frequently two closely associated single papillae instead of the double papilla at one side, as shown in Fig. 6), and two pairs of single papillae posteriorly. A phasmidial pore was located between the last two papillae on each side. This postcloacal papillar arrangement was similar to that of A. lumbricoides, except for the position of the phasmidial pores (Fig. 27), but was distinctly different from that of T. leonina, as shown in Fig. 17. In the female tail of the three species, only one pair of phasmidial pores was recognized, situated laterally, near the terminal end (Fig. 9). Terminal ends of both male and female specimens of all three species usually exhibited a pointed spiny formation (Figs. 8, 9, 19), but blunt or button-like tips were noted exceptionally, as shown in Fig. 27.

The Egg

Eggs from *B. transfuga* showed an uneven, crepuscular surface in high magnification (Fig. 11), and under light microscopy we believe the operculum-like area was probably there, as seen in one pole of the egg shell in some eggs. In *A. lumbricoides* the egg showed distinct depressions and ridges all over the surface except one small area, where it appears differently at one end, probably corresponding to the operculum-like structure by Ubelaker and Allison (1975) (Fig. 26). In *T. leonina* the egg surface was smooth (Fig. 18).

Discussion

SEM of the surface structures of Ascaris species and Baylisascaris species had been reported in part by various researchers (Weise, 1973; Sprent et al., 1973; Kurimoto, 1974; Kikuchi et al., 1979).

In this report we have included some new findings through SEM examinations of these species and have made a comparison of the SEM of the species on the basis of our present results. The surface of the subventral and dorsal labial papillae of *B. transfuga* was more similar to that of

T. leonina than to that of A. lumbricoides. The surfaces of the lateral papillae were similar among the species which were included in the subfamily Ascaridinae, but different from those of the Toxocara species (Uni and Takada, 1975). The cervical alae could be recognized vestigially in A. lumbricoides, but here the cuticular bars were distinctly different from those of B. transfuga (Uni et al., 1980a) and T. leonina as revealed in the previous examination (Uni et al., 1980b). With regard to the pericloacal rough area, it was revealed in A. lumbricoides as being vestigial. Our present findings confirmed the probability of the operculum-like structure of the eggs of A. lumbricoides.

Through our SEM examination we found that *B. transfuga* showed close morphological affinity to both *A. lumbricoides* and *T. leonina* in various parts of the body. The detailed SEM comparison of particular points should be useful in clarifying the affinities and differences between the species of the subfamily Ascaridinae.

Summary

The external structures of adult worms and eggs of species *B. transfuga*, *T. leonina* and *A. lumbricoides* were examined under SEM. Cervical alae were found vestigially in the specimens of *A. lumbricoides*. The unique pericloacal structure of *B. transfuga* was clearly visualized, and the corresponding structure could be found vestigially in *A. lumbricoides*. The surface structures of the three species were compared.

Acknowledgement

We express our appreciation to Mr. M. Miyashita, Tennoji Zoological Gardens, Osaka, for the donation of the specimens from bears.

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Baylisascaris transfuga, イヌ小蛔虫 (T. leonina) および 蛔虫 (A. lumbricoides) の走査電子顕微鏡による表面構造の比較研究

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B. transfuga, イヌ小蛔虫, 蛔虫の雌雄成虫, 虫卵 の表面構造を走査電子顕微鏡を用いて調べた. 頭部の lateral papilla は三種において極めて類似していた. B. transfuga の亜腹側口唇乳頭 (subventral labial papilla) および背側口唇乳頭 (dorsal labial papilla) はイヌ小蛔虫のそれに類似していたが, 蛔虫とは異な っていた. 蛔虫の頸部において頸翼が, 痕跡的である が, 見い出された. 三種の頸翼の背側基部に頸乳頭 (deirid) が位置していた. Baylisascaris の属標徴であ る pericloacal rough area の詳細が明らかになり, それに相当する構造物が蛔虫においても、痕跡的であ るが、見い出された. 雄虫体尾部の乳頭の配列は B. transfuga および 蛔虫において互いに極めて類似して いたが、ファスミッドの位置は異なっていた. 蛔虫卵 において卵蓋様構造物と考えられるものが 認められ た. これらの知見より B. transfuga は 蛔虫およびイ ヌ小蛔虫と、その虫体の異なった部位において、極め て類似した構造を示す様に考えられる.

Explanation of Figures

- Figs. 1-11 Baylisascaris transfuga.
- Fig. 1 En face view of head of female. The area marked with * is enlarged in Fig. 4. ×100.
- Fig. 2 Denticles in central area of subventral lip. ×2,000.
- Fig. 3 Amphidial pore and lateral papilla. ×1,000.
- Fig. 4 Dorsal labial papilla. ×1,000.
- Fig. 5 Deirid. ×2,000.
- Fig. 6 Ventral view of male posterior part. * shows posterior part of pericloacal rough area. ×150.
- Fig. 7 Pericloacal rough area and precloacal central papilla. ×200.
- Fig. 8 Lateral view of male tail. $\times 400$.
- Fig. 9 Posterior part of female. ×100.
- Fig. 10 Egg. ×350.
- Fig. 11 Surface of egg. ×5,600.
- Figs. 12-20 Toxascaris leonina.
- Fig. 12 En face view of head of male. The area marked with * is enlarged in Fig. 15. ×200.
- Fig. 13 Denticles in central area of dorsal lip. $\times 2,000$.
- Fig. 14 Amphidial pore and lateral papilla. $\times 1,400$.
- Fig. 15 Dorsal labial papilla. $\times 1,400$.
- Fig. 16 Deirid. ×1,800.
- Fig. 17 Ventral view of male posterior part. The area marked with * is enlarged in Fig. 19. $\times 200$.
- Fig. 18 Egg. ×350.
- Fig. 19 Lateral view of male tail. ×500.
- Fig. 20 Area around cloaca. ×400.
- Figs. 21-28 Ascaris lumbricoides.
- Fig. 21 En face view of head of female. The area marked with * is enlarged in Fig. 24. $\times 100$.
- Fig. 22 Denticles in central area of dorsal lip. ×2,000.
- Fig. 23 Amphidial pore and lateral papilla. ×1,000.
- Fig. 24 Dorsal labial papilla. ×700.
- Fig. 25 Cervical ala and deirid. ×350.
- Fig. 26 Presumptive operculum-like structure (*) of egg. $\times 3,500$.
- Fig. 27 Ventral view of male posterior part. * shows rough area. ×200.
- Fig. 28 Phasmidial pore. \times 3,500.
 - Abbreviations
 - A: anus
 - AM: amphidial pore
 - CA: cervical ala
 - D: deirid
 - IP: internal labial papilla
 - LP: lateral papilla
 - PCP: precloacal central papilla
 - PH: phasmidial pore
 - S: spicule

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