INTRODUCTION

Nematode parasites of the genus Thelazia are clinically important, causing insect-borne zoonoses in humans. More than 10 species have been reported in various parts of the world. Among them, Thelazia callipaeda, the oriental eye worm, is widespread in domestic and wild animals in Asian countries, Italy, France, and Germany, and it is sometimes found in humans in China, India, Thailand, Indonesia, Japan, and Korea [1-4]. In the Republic of Korea, a total of 37 human infection cases were reported in the literature since the first one by Nakada (1934) [5-32]. Recently, we found 2 cases of human thelaziasis (HT) that occurred in Gyeongsangnam-do, and we analyzed characteristics of the overall 39 Korean HT cases, including the present 2 cases.

CASE RECORD

Case 1

A 58-year old woman, residing in Hadong-gun, Gyeongsangnam-do, came to Gyeongsang National University Hospital (GNUH) complaining of foreign body sensation and itching of the right eye in March 2000. Total 6 adult nematodes of Thelazia callipaeda (2 males and 4 females) were detected in her right eye. A 80-year old man, residing in Jinju-si, Gyeongsangnam-do, came to GNUH complaining of foreign body sensation, itching, and pain of the right eye in December 2007. A total of 5 worms (4 females and 1 degenerated) were removed from his right eye. We analyzed characteristics of the total 39 Korean HT cases reported to date, including the present 2 cases. Most of the cases (71.8%) occurred in Seoul and Gyeonggi-do before 2000, and 21 cases (53.8%) were males and 18 (46.2%) were females. The prevalence was higher in younger ages below 30 years (48.7%) than 31-60 years (41.0%) and over 61 years (10.3%). The seasonal prevalence showed a higher incidence in autumn (43.6%) than in other seasons. Most of the cases (94.9%) were conjunctival sac infections and only 2 (5.1%) were intraocular cases. The present 2 HT cases are the first reported cases in Gyeongsangnam-do. Some characteristics of Korean HT cases were analyzed.
moved by a physician in a local hospital in Jinju-si, Gyeongsangnam-do. A total of 4 worms (3 females and 1 dead and degenerated) were extracted from his right eye. At 1 month later, 1 female worm was also removed from his right eye at GNUH.

DESCRIPTION OF WORMS

Among 10 worms collected, 6 females and 1 male were fixed with 10% hot formalin, cleared in alcohol-glycerin solution, mounted in glycerin-jelly, and observed under a light microscope (LM) with a micrometer. To observe the surface ultrastructure, remainders, 2 females and 1 male, were washed several times with 0.2 M cacodylate buffer (pH 7.2) and fixed with 2.5% glutaraldehyde at 4°C. After washing 3 times with the buffer, they were dehydrated through a graded alcohol series (50%, 70%, 80%, 90%, 95%, and absolute alcohol), dried with hexamethyldisilazane, coated (JFC-1100E ion sputtering device) with gold, and observed with a scanning electron microscope (Philips XL-30S, Einhoven, The Netherlands) at an accelerating voltage of 20 kV.

The male worm was 12.4 × 0.44 mm in size, had a scalariform buccal cavity (0.025 × 0.033 mm), a long muscular esophagus (0.65 mm long), and a coiled tail. In the scanning electron microscopic (SEM) study, the cephalic part (about 0.028 × 0.044 mm) with the buccal cavity, 2 cephalic papillae, and a body papilla which located at anterior 1/10 in a longitudinal groove, were characteristically observed in the anterior portion. Folded transverse striations were arranged on the anterior surface from just behind the cephalic part. Their densities were about 375 rows per 1 mm length of the cuticle (Fig. 2). Cuticular transverse striations arranged in the middle and posterior portions were about 220 and 240 rows per 1 mm length, respectively. A spicule and dome-shaped papillae, 7 pairs of preanal, and 2 pairs of postanal papillae were observed in the coiled tail part (Fig. 3).

Females were 17.0-18.5 × 0.39-0.49 mm (17.7 × 0.44 mm on average) in size, had a scalariform buccal cavity (0.028 × 0.033 mm), a long muscular esophagus (0.70 mm long) and a conical tail. Vulva openings were located at 0.52-0.6 mm (0.57 mm on average) from the anterior end and at more anterior portion than the esophago-intestinal junction. Numerous coiled larvae were contained in the proximal part of the uterus, and lots of eggs were present in the distal part of the uterus. In the SEM study of the body surface, folded transverse striations arranged on the anterior surface were about 250 rows per 1 mm length. Cuticular transverse striations arranged in the middle and posterior portions were about 170 and 375 rows per 1 mm length, respectively. Encircled striations were distributed on the posterior end (Fig. 4).

DISCUSSION

In the present study, 2 cases of HT were reported for the first time in Gyeongsangnam-do. Therefore, including the present
2 cases, a total of 39 cases of HT have been documented in the Republic of Korea. Among them, 28 cases (71.8%) occurred in Seoul and Gyeonggi-do (Province), and the remainders (11 cases) occurred in Chungcheongbuk-do (2 cases), Jeollabuk-do (1 case), Daegu (1 case), Gyeongsangnam-do (2 cases), Busan (1 case), and other areas (3 cases). It is interesting to note that all 5 cases reported from Southeastern parts (Gyeongsangbuk-do, Daegu, Gyeongsangnam-do, and Busan) occurred after 2000, whereas all cases of Seoul and Gyeonggi-do occurred before 2000 (Table 1). The plenty of HT cases in Seoul and Gyeonggi-do may be related to an abundance of academic facilities and manpowers which can aware and report the parasitic disease, as well as to the increase of outdoor activities of residents. However, it is meaningful to see that HT cases are now being reported chiefly in Southeastern parts after 2000. In China, a total of 371 HT cases had been reported by 2005, and most of them occurred in rural populations unlike in Korea [4].

Total 3 cases were reported before the 1960s. Among them, 2 cases occurred in the Northern part of Korea, Hwanghae-do and Pyeonganbuk-do [5-7]. There were no reports during the 1960s, 6 (15.4%) in the 1970s, 11 (28.2%) in the 1980s, 13
(33.3%) in the 1990s, and 6 (15.4%) after 2000. A total of 21 (53.8%) cases were males and 18 (46.2%) were females. By age groups, 19 cases (48.7%) occurred below 30 years of age, 16 cases (41.0%) in 31-60 years, and 4 cases (10.3%) at over 61 years (Table 1). On the other hand, a review on the analysis of 179 Chinese HT cases revealed 51% from females and 49% from males, and showed that the majority (64.2%) of patients were children younger than 6 years [4].

Seasonal prevalences were clearly shown in the cases of Korean HT. The highest incidence of 17 cases (43.6%) occurred in autumn (6 cases in September: 5 in October, and 6 in November). In the summer season, total 7 (17.9%) cases (1 case in June, 3 in July, and 3 in August) occurred. A total of 5 (12.8%) and 3 (7.7%) cases were encountered in winter (4 cases in December and 1 in January) and spring (each 1 case in March, April, and May). There was no case in February, and the occurrence date was unknown in 7 (17.9%) cases. The prevalence (69.2%) of the second half year, from July to December, is much higher than that (12.8%) of the first half year, from January to June. Therefore, seasonal tendency may be related to increasing outdoor activities of humans and increase of the vector population.

A total of 146 worms (44 males, 62 females, and 40 unknown) were recovered from all 39 Korean cases. The individual worm burden was 1-15 worms per patient (3.7 on average). In the case of the maximum worm burden (15 worms), the infection occurred at 25 days after the primary worm recovery (10 worms) [31]. Kim and Shin [24] removed 14 worms at once from the right eye of a 7-year old girl in July 1989. A total of 21 cases (52.5%) had infection in the right eye, 16 (40.0%)}
Two cases of thelaziasis in Korea

in the left eye, 3 (7.5%) unknown, and only 1 case had infection in both eyes. Only in 2 cases (5.1%), worms were found in the intraocular sites, whereas in the most of cases (94.9%), worms were detected in the conjunctival sac [30,32].

The chief complaints of Korean cases with *T. callipaeda*, especially conjunctival sac infections, were foreign body sensation (78.8%), itching (48.5%), and lacrimation (36.4%). They sometimes complained of hyperemia and hemorrhage of the eyes, mild visual disturbances, blurring sensation, and pain as the minor symptoms. The visual disturbance, floaters, myodesopsia, and vitreous opacity were manifested in intraocular cases [30,32].

For diagnosis of HT, identification of worms extracted from the eye is important. Therefore, many researchers observed morphological characteristics, including surface ultrastructure, of worms recovered from human cases [21,23,27,33-35]. We also examined the morphologies of *T. callipaeda* worms recovered from a case to provide more clear and profitable information in this study. Internal structures, such as the scalariform buccal cavity, muscular esophagus, esophageal nerve ring, intestine, rectum, anus, spicule, vulva opening, vagina, and larvae and eggs in uteri, were well visualized in light microscopic

| Table 1. Analytical summery of thelaziasis cases reported in the Republic of Korea |
|-------------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| Items             | Before the 1960’s | 1970’s          | 1980’s          | 1990’s          | 2000-present    |
| Sex               |                 |                 |                 |                 |                 |
| Male              | 2               | 3               | 6               | 9               | 1               |
| Female            | 1               | 3               | 5               | 4               | 5               |
| Total (%)         | 3 (7.7)         | 6 (15.4)        | 11 (28.2)       | 13 (33.3)       | 6 (15.4)        |
| Age               |                 |                 |                 |                 |                 |
| Below 20          | 3               | 2               | 1               | 2               | 1               |
| 21-30             | -               | 4               | 4               | 2               | -               |
| 31-40             | -               | -               | 3               | 1               | -               |
| 41-50             | -               | -               | 2               | 4               | -               |
| 51-60             | -               | 1               | 3               | 2               | 6               |
| 61-70             | -               | -               | -               | 1               | -               |
| Over 71           | -               | -               | -               | 3               | 3               |
| Total             | 3               | 6               | 11              | 13              | 6               |
| Residential district of cases |                 |                 |                 |                 |                 |
| Seoul             | 1               | 6               | 5               | 5               | -               |
| Gyeonggi-do       | -               | -               | 5               | 6               | -               |
| Chungcheongbuk-do | -               | -               | -               | 2               | -               |
| Jeollabuk-do      | -               | 1               | -               | -               | -               |
| Gyeongsangbuk-do  | -               | -               | -               | -               | 1               |
| Daegu             | -               | -               | -               | -               | 1               |
| Gyeongsanam-do    | -               | -               | -               | -               | 2               |
| Busan             | -               | -               | -               | -               | 1               |
| Others            | 2a              | -               | -               | -               | 1b              |
| Total             | 3               | 6               | 11              | 13              | 6               |
| Eyes infected     |                 |                 |                 |                 |                 |
| Right             | -               | 3               | 5               | 10              | 3               |
| Left              | 1               | 3               | 7               | 2               | 3               |
| Unknown           | 2               | -               | -               | 1               | -               |
| Total             | 3               | 6               | 12              | 13              | 6               |
| No. of worms detected |                 |                 |                 |                 |                 |
| Male              | 1               | 5               | 11              | 20              | 7               |
| Female            | 1               | 3               | 20              | 20              | 18              |
| Unknown           | 1               | 5               | 7               | 20              | 7               |
| Total             | 3 (2.1)         | 13 (8.9)        | 38 (26.0)       | 60 (41.1)       | 32 (21.9)       |

*a* Two cases occurred in Northern parts of Korea; *b* Unknown; *c* One case was infected in both eyes.
observations. Surface ultrastructures, including the cephalic part, cephalic papillae, body papilla, transverse striations by body portions, spicule, and preanal and postanal papillae, were well revealed by the SEM study. The morphological findings observed in this study were well coincided with those of the previous studies. However, the fine and clear LM and SEM images obtained in this study will be helpful to identify *T. callipaeda* from HT cases [21,23,27,33-35].

Fig. 4. LM (A-D) and SEM views (E-H) of *T. callipaeda* females. (A) Anterior portion with the scalariform buccal cavity (BC), a long muscular esophagus (E), and a vulva opening (VO) which is located at more anterior portion than the esophago-intestinal junction (arrow) (scale bar = 1 mm). (B) The proximal part of the uterus with numerous coiled larvae (scale bar = 1 mm). (C) The distal part of the uterus with lots of eggs (scale bar = 1 mm). (D) The posterior end of a worm with the rectum (R) and anus (A) (scale bar = 1 mm). (E) The anterior portion with folded transverse striations, which were arranged about 250 rows per 1 mm length. (F) The middle portion with transverse striations, which were arranged about 170 rows per 1 mm length. (G) The posterior portion with transverse striations, which were arranged about 375 rows per 1 mm length. (H) The posterior end with encircled striations (scale bar = 10 µm).

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