

# The Jeju Weasel, *Mustela sibirica quelpartis*, A New Definitive Host for *Gnathostoma nipponicum* Yamaguti, 1941

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**Abstract:** Adult gnathostomes were discovered in the stomach of the Jeju weasel, *Mustela sibirica quelpartis*, road-killed in Jeju-do (Province). Their morphological characters were examined to identify the species. Total 50 gnathostome adults were collected from 6 out of 10 weasels examined. In infected weasels, 4-6 worms were grouped and embedded in each granulomatous gastric tumor, except 1 weasel. Male worms were 25.0 × 1.4 mm in average size, and had a tail with pedunculate papillae, a spicule, and minute tegumental spines. Females were 40.0 × 2.5 mm in average size, and had a tail without tegumental spines. Pointed and posteriorly curved hooklets were arranged in 8-10 rows on the head bulb. Tegumental spines were distributed from behind the head bulb to the middle portion of the body. The spines were different in size and shape by the distribution level of the body surface. Fertilized eggs were 65.5 × 38.9 μm in average size, and had a mucoid plug at 1 pole. These gnathostomes from Jeju weasels were identified as *Gnathostoma nipponicum* Yamaguti, 1941. By the present study, it was confirmed for the first time that *G. nipponicum* is distributed in Jeju-do, the Republic of Korea, and the Jeju weasel, *M. sibirica quelpartis*, plays a crucial role for its definitive host.

**Key words:** *Gnathostoma nipponicum*, Jeju weasel, *Mustela sibirica quelpartis*, new definitive host

Nematode parasites of the genus *Gnathostoma* are clinically important, causing food-borne zoonoses in humans. More than 10 species have been reported in various parts of Asia, Europe, Oceania, and Americas. In the Republic of Korea, there had been no reports on gnathostome infections before a paper by Kim in 1973 [1]. He detected 2 *Gnathostoma spinigerum* larvae in the abdominal muscle of a snakehead, *Channa argus argus*, from Kimhae, Gyeongsangnam-do (Province). After that time, third stage larvae of *Gnathostoma nipponicum* and *Gnathostoma hispidum* were found in the imported Chinese loaches [2,3]. In addition, *G. hispidum* larvae were detected in pit-viper snakes, *Agkistrodon brevicaudus*, caught in Korea, and also in red banded odd-tooth snakes, *Dinodon rufozonatum rufozonatum*, from China [4,5]. However, adult gnathostomes had never been reported. In the present study, we found adult gnathostomes in

the stomach of the Jeju weasel, *Mustela sibirica quelpartis*, road-killed in Jeju-do (Province), the Republic of Korea, and identified them as *G. nipponicum* on the basis of their morphological characters.

A total of 10 Jeju weasels, *M. sibirica quelpartis*, road-killed, were donated from the party of environmentalists of Jeju-do from April 2008 to April 2010 (Fig. 1). Their whole visceral organs were isolated to investigate parasite infections. Adult nematodes were collected from the stomach of the weasels. Some collected worms were fixed with 10% formalin, cleared in alcohol-glycerin solution, mounted in glycerin-jelly, and observed under a light microscope. To observe the surface ultrastructure, some worms 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, Eindhoven, The Netherlands) at an accelerating voltage of 15 kV.

Total 50 gnathostomes were collected from 6 out of 10 wea-

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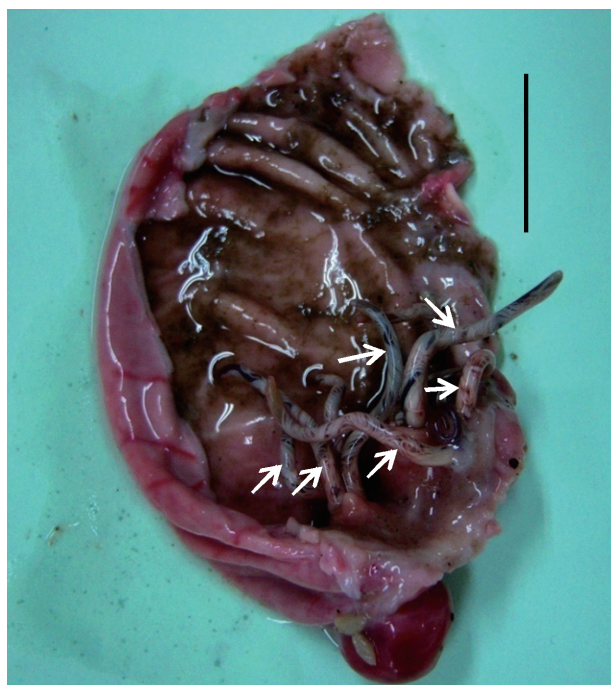
sels examined, and they were mainly from weasels caught in Jeju-si (Table 1). Except in 1 weasel, 4-6 worms were grouped and embedded in each granulomatous gastric tumor of weasels (Fig. 2). Pointed and posteriorly curved hooklets were arranged in 8-10 rows on the head bulb (Fig. 3A, 3B). Male worms were 22.0-27.0×1.2-1.9 (25.0×1.4) mm in size, and had a characteristic head bulb and a tail with pedunculate papillae, a spicule, and minute tegumental spines (Fig. 3C, 3D). Females were 35.0-45.0×2.3-2.8 (40.0×2.5) mm in size, and had a head bulb and a tail without tegumental spines. Fertilized eggs were 63.8-67.5×37.5-40.0 (65.5×38.9)  $\mu\text{m}$  in size, and had a mucoid plug at 1 pole (Fig. 4). Tegumental spines were distributed only on the anterior half of the body. They were different in size and shape by the distribution level of the body surface. Slender 3-4 tipped spines (31.4×11.6  $\mu\text{m}$  in average size) were



**Fig. 1.** The Jeju weasel, *Mustela sibirica quelpartis*, a new definitive host for *Gnathostoma nipponicum*, which was photographed by Hong-Shik Oh, one of the authors.

densely distributed just behind of the head bulb (Fig. 5A). Broader 3 tipped spines (44.6×18.3  $\mu\text{m}$  in average size) with a long middle tip (25.0  $\mu\text{m}$  in average length) were broadly distributed at the anterior half of the middle surface. Slender 3 tipped ones (34.0×11.8  $\mu\text{m}$  in average size) and 2-3 tipped small pointed spines (about 20  $\mu\text{m}$  in length) were distributed on the posterior half of the middle surface (Fig. 5B, 5C, 5D). No tegumental spines were distributed on the posterior half of the body surface, especially in females (Fig. 5E). In males, minute tegumental spines were distributed only at the tail portion.

It has been known that *G. nipponicum* is a common parasite



**Fig. 2.** Adult worms of *G. nipponicum* (arrow marks) embedded in the granulomatous gastric tumor of a weasel. Scale bar = 2 cm.

**Table 1.** The road-killed Jeju weasel, *Mustela sibirica quelpartis*, examined and the result of worm recovery

Weasel no.	Weight (g) of weasels	Date examined	Location of road-killed	No. of <i>G. nipponicum</i> worms recovered
1	UD <sup>a</sup>	4 April, 2008	Odung-dong, Jeju-si	9
2	242	30 August, 2008	Gumak-ri, Bukjeju-gun	0
3	307	20 September, 2008	Ara-dong, Jeju-si	0
4	528	28 September, 2008	Gumak-ri, Bukjeju-gun	0
5	UD <sup>a</sup>	16 October, 2008	Gujoa-ri, Namjeju-gun	0
6	669	15 November, 2008	Goirai-ri, Jeju-si	12
7	551	14 March, 2009	Goirai-ri, Jeju-si	10
8	597	21 April, 2009	Goirai-ri, Jeju-si	9
9	561	14 April, 2010	Cheonwang-san, Jeju-si	1
10	302	24 April, 2010	Jeju-si	9

<sup>a</sup>Undetermined.

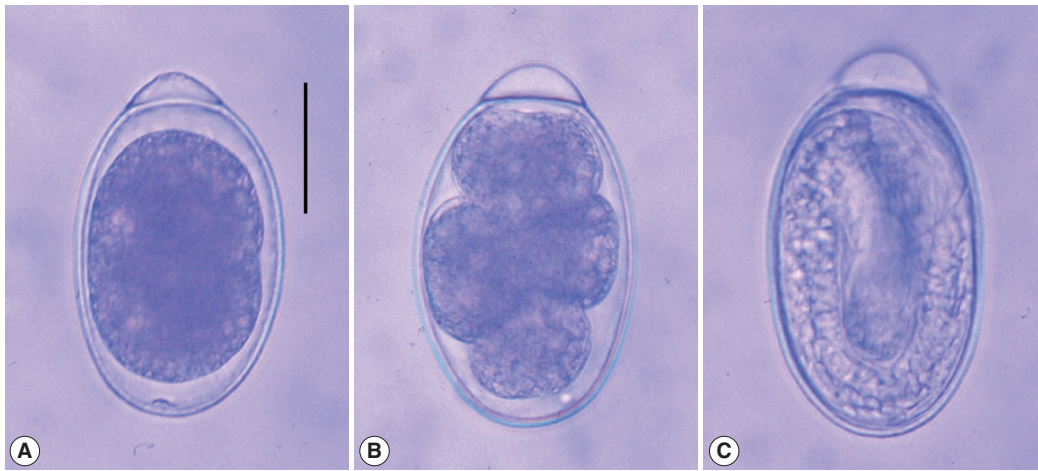


**Fig. 3.** Morphological characters of *G. nipponicum* adults recovered from a Jeju weasel. Light (A) and scanning electron microscopic (SEM) (B) views of the head bulb with 10 rows of hooklets. Light (C) and SEM (D) views of the tail portion of a male with pedunculate papillae, a spicule, and minute tegumental spines. Scale bar = 0.2 mm.

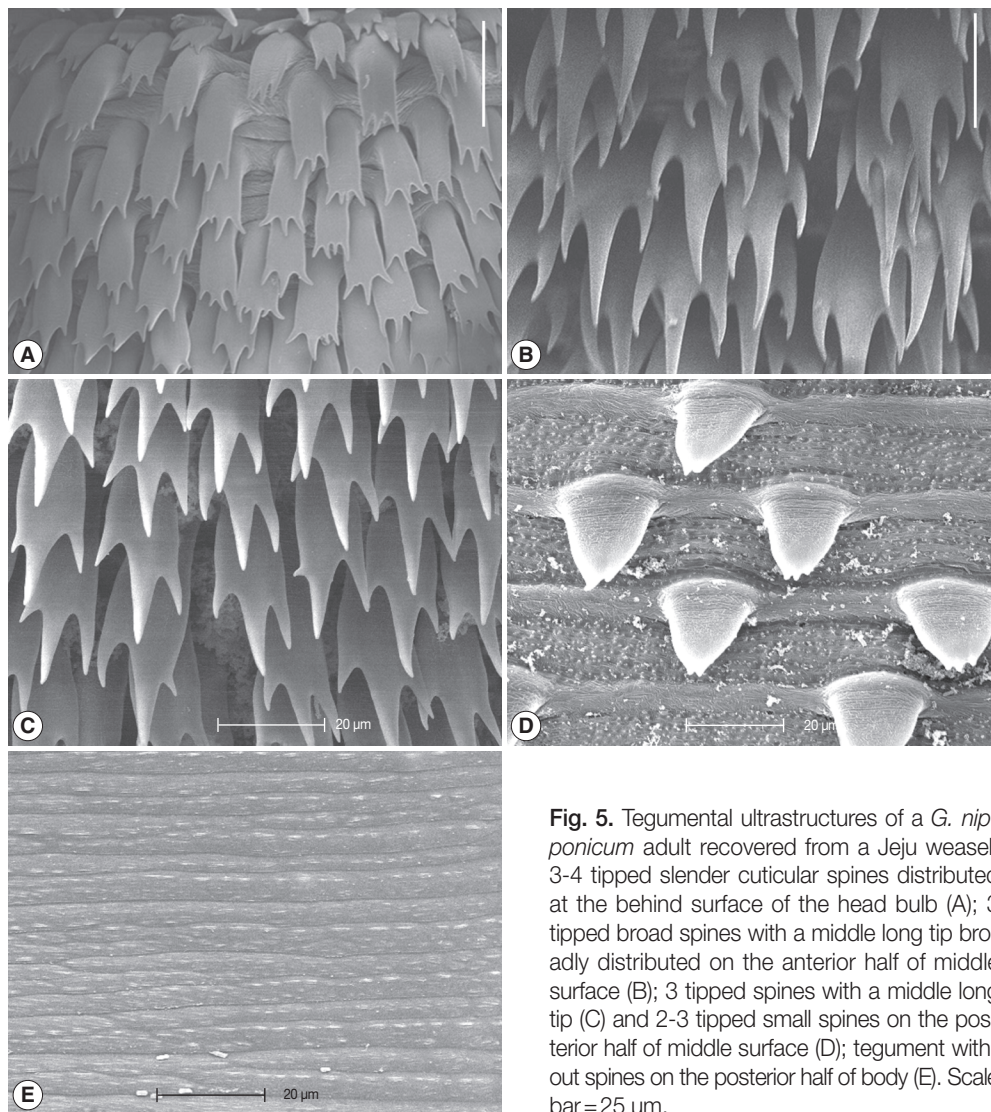
found in esophageal tumors of weasels, *Mustela sibirica itasi* and *Mustela sibirica coreana*, only in Japan. This nematode was first found by Yoshida in 1931 [6] in the esophagus of a weasel from the middle part of Honshu in Japan, and erroneously named as *G. spinigerum*. Subsequently, by many Japanese workers, it was revealed that this parasite is widely distributed in more than 23 Prefectures in Japan [7-15]. By the present study, it was

confirmed for the first time that *G. nipponicum* is distributed also in Jeju-do, the Republic of Korea, and the Jeju weasel, *M. sibirica quelpartis*, plays a crucial role for a definitive host.

Detailed morphological features of adult *G. nipponicum* have been described by Yamaguti in 1941 and later by Miyazaki and Umetani in 1950 [9,16]. Miyazaki [17] reviewed 10 valid species of *Gnathostoma*, i.e. *G. spinigerum*, *G. hispidum*, *G. doloresi*, *G.*



**Fig. 4.** An egg of *G. nipponicum* collected from the uterus of an adult female (A). Developing eggs from the cultivating petri dish with freshwater at 28°C are seen (B & C). Scale bar = 25 µm.



**Fig. 5.** Tegumental ultrastructures of a *G. nipponicum* adult recovered from a Jeju weasel. 3-4 tipped slender cuticular spines distributed at the behind surface of the head bulb (A); 3 tipped broad spines with a middle long tip broadly distributed on the anterior half of middle surface (B); 3 tipped spines with a middle long tip (C) and 2-3 tipped small spines on the posterior half of middle surface (D); tegument without spines on the posterior half of body (E). Scale bar = 25 µm.

*nipponicum*, *G. turgidum*, *G. procyonis*, *G. miyazakii*, *G. malaysiae*, *G. vietnamicum*, and *G. americanum* [17]. According to this review [17], the distribution and shape of tegumental (cuticular) spines on the body surface were treated as the most useful landmarks for species differentiation of *Gnathostoma*. In 6 species, *G. spinigerum*, *G. nipponicum*, *G. turgidum*, *G. miyazakii*, *G. vietnamicum*, and *G. americanum*, their tegumental spines are covered at the anterior half of the body, while in remaining 4 species, *G. hispidum*, *G. doloresi*, *G. procyonis*, and *G. malaysiae*, the tegumental spines are densely distributed on the whole body surfaces. However, *G. nipponicum* can be discriminated from other 5 species, including *G. spinigerum* by the morphology of tegumental spines and eggs. Broader 3 tipped spines with a long middle tip, which are broadly distributed at the anterior half of the middle surface, are most characteristic and comparable features for species identification. Additionally, the tegumental ultrastructures of adult *G. nipponicum* are clearly shown by the present study.

It has been known that *G. nipponicum* adults are found in esophageal tumors of weasels, *M. sibirica itasi* and *M. sibirica coreana*, while most of other *Gnathostoma* spp. are usually found in the stomach of definitive hosts, including cats, dogs, and pigs [17]. However, in the present study, the worms were detected in the stomach of Jeju weasels, *M. sibirica quelpartis*. It is interesting that habitats of *G. nipponicum* in the definitive hosts are different between weasels from Japan and from Korea. Molecular genetic studies may be needed to know what is differential points between Japanese and Korean worms, although they are morphologically identical.

**Conflict of interest:** The authors have no conflicts of interest concerning the work reported in this paper.

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