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A Survey on *Gnathostoma hispidum* Fedtschenko, 1872 in Jiangxi, People's Republic of China

WEN CHANG HUANG[1], BAO FEN XIA[1], MASATAKA KOGA[2] AND YOICHI ISHII[2]

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Morita *et al.* (1984) reported 8 cases of gnathostomiasis between 1980 and 1981 in western Japan. The patients had eaten live loaches, *Misgurnus anguillicaudatus* which were believed to have been imported from People's Republic of China, Taiwan or Korea. Symptoms included creeping eruptions and eosinophilia. Examination of larvae recovered from loaches revealed a small type, less than 1 mm long, and a large type, about 3 mm long. Most of the larvae obtained were of the small type (99 %). The small larvae were successfully transmitted to pigs and later identified as *Gnathostoma hispidum* (Akahane *et al.*, 1982; Koga *et al.*, 1984; Takakura *et al.*, 1985). The large larvae could not be transmitted to pigs or other animals and have not been identified. Until recently there have been no reports on the existence of *G. hispidum* in Japan.

The life cycle of gnathostomes can be produced experimentally. Sheathed larvae hatch from eggs and move actively in fresh water. They are ingested by cyclops and move to the pseudocoelomic cavity of the invertebrates (the first intermediate host). The larvae molt twice to become early third stage larvae ($E.L_3$). The $E.L_3$ are about 0.4–0.7 mm in body length and bear head bulbs. The $E.L_3$ grow to a size of about 3.0–4.0 mm and become advanced third stage larvae ($Ad.L_3$) after transmission to fishes, frogs, birds or mammals (the second intermediate or paratenic hosts). These larvae are able to become adults in the final hosts. The adult worms and $Ad.L_3$ of *G. hispidum* are the only stages of this parasite that have been recovered from field collections of vertebrates. And the first intermediate hosts harboring the $E.L_3$ in nature are still unknown. The small larvae, measuring 0.6–1.0 mm, were found in naturally infected loaches and resembled $E.L_3$. The $E.L_3$ might occur in fishes as well as in copepods. To help elucidating the life cycle of *G. hispidum* under natural conditions, the presence of adult worms was examined in Jiangxi Province in mainland China where the parasite is thought to be endemic.

1) *Department of Parasitology, Jiangxi Medical College, Nanchang, People's Republic of China*
2) *Department of Parasitology, Faculty of Medicine, Kyushu University, Fukuoka 812, Japan*

Materials and Methods

Eleven regions were randomly chosen in Jiangxi Province – Gaoan, Fengxin, Jishui, Linchuan, Shanggao, Fengcheng, Nanfeng, Nanchang, Xingan, Xiajiang and Yongxiu. We examined the stomachs of 844 pigs (Chinese Breed: Bin Hu Hei Zhu, Gan Zhong Nan Hua Zhu, Liang Tou Wu Zhu, Gan Dong Hei Zhu) at slaughter houses in these regions. Stomachs were carefully opened along their lesser curvatures by dissecting scissors. Adults of *G. hispidum* were usually found in shallow tumors on the stomach wall and could be easily detached. The contents of each stomach were also examined.

Worms and eggs were washed in physiological saline and fixed in 10% formalin. Some were processed for scanning electron microscopy (SEM). Adult males and females were soaked in running water overnight to remove formalin. They were subsequently rinsed in Millonig's phosphate buffer and postfixed in 1% OsO_4 for two hours. The worms were cut transversely into four parts during postfixation to facilitate observations by SEM. They were then dehydrated in a graded series of ethanol, transferred into amyl acetate and dried in liquid CO_2 using a Hitachi HCP-2 critical-point dryer. The specimens were coated with gold in an ion-sputtering apparatus (JEOL FC-1100) and observed under a JEOL JSM-U3 scanning electron microscope operated at 15 kV. Eggs were processed in the same manner.

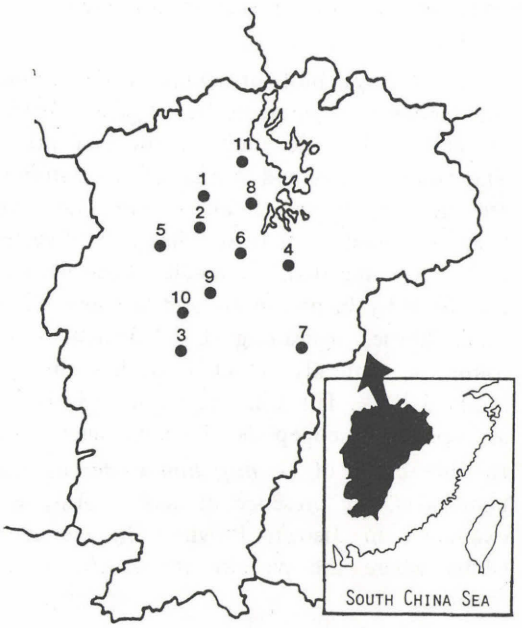


Fig. 1 Map showing the locations chosen to investigate the fauna of *G. hispidum* in Jiangxi Province. 1. Gaoan 2. Fengxi 3. Jishui 4. Linchuan 5. Shanggao 6. Fengcheng 7. Nanfeng 8. Nanchang 9. Xingan 10. Xiajiang 11. Yongxiu

Results

Jiangxi Province is located in south-central part of mainland China. Nanchang is the main city of this province. All regions surveyed were positive for gnathostomes except for Fengxin (Fig. 1) (Table 1). The prevalence ranged from a high of 13.6% in Nanchang to a low of 3.4% in Gaoan. Totally sixty seven

Table 1 Recovery of *G. hispidum* from pigs at slaughter houses in Jiangxi Province

Locality	No. Examined	No. Infected	Infection Rate (%)
1. Gaoan	29	1	3.4
2. Fengxin	23	0	0.0
3. Jishui	20	1	5.0
4. Linchuan	56	4	7.1
5. Shanggao	77	5	6.7
6. Fengcheng	63	6	9.5
7. Nanfeng	51	5	9.8
8. Nanchang	125	17	13.6
9. Xingan	80	5	6.3
10. Xiajiang	215	12	5.6
11. Yongxiu	105	11	10.4
Total	844	67	7.9

Fig. 2 Anterior view of adult *G. hispidum* with head bulb armed with 11 transverse rows of hooks. ×85

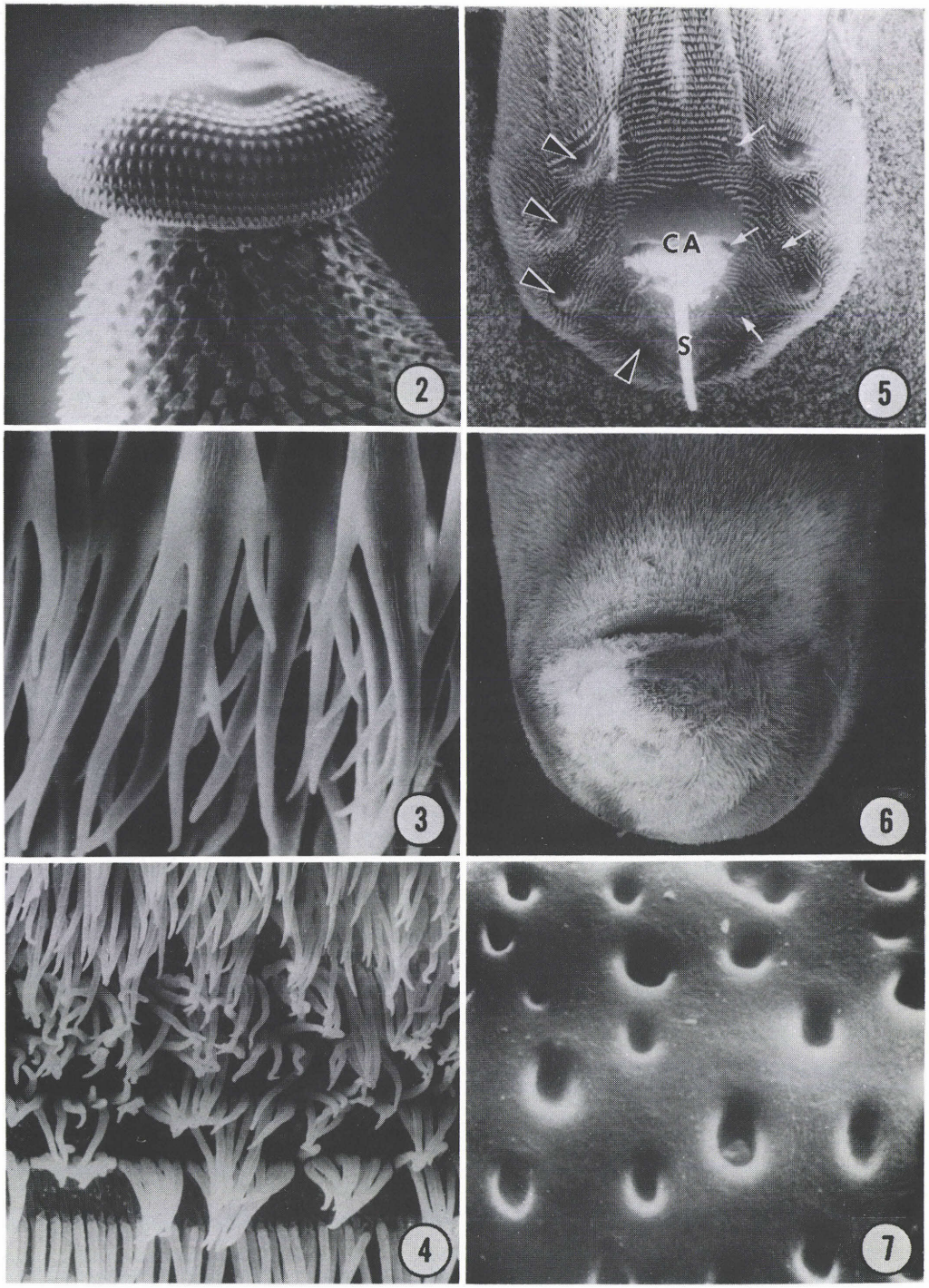
Fig. 3 Typical three denticle spines of *G. hispidum*. The middle denticle is markedly longer than the lateral two. ×750

Fig. 4 Single pointed short spines cover the posterior part of the body. ×750

Fig. 5 Ventral surface of a male terminal end covered wholly with small spines. Black arrows indicate caudal papillae. White arrows indicate small papillae. S: spicule CA: cloacal papillae. ×100

Fig. 6 A female tail extremity covered densely with minute cilia-like spines. ×80

Fig. 7 Egg shell surface having comparatively deeper pits. ×7,500


2
CA
S
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of 844 pigs (7.9%) were positive for adult gnathostomes.

When observed by SEM, the worms had hemispherical head bulbs, armed with 9–12 rows of hooks (Fig. 2). Cuticular spines were present throughout the length of the worms along their transverse striations. The spines varied greatly in size and shape in various body regions. The largest variation occurred in the anterior third of the body. Spines immediately behind the head bulb were small and stumpy and bore five to ten teeth. Posterior to these, the spines were longer and had three teeth. The middle tooth was markedly elongated (Fig. 3). Posterior to the three-toothed spines were spines with two denticles. Regions having two- and three-toothed spines were very narrow. On the posterior two-thirds of the body, the spines were single-pointed. The spines gradually became short in length towards the posterior end of the worm (Fig. 4). The ventral surface of the posterior end of male worms had four pairs of caudal papillae and three pairs of small papillae (Fig. 5, arrows) and was covered with small unidentate spines. The spines were absent around the papillae. The posterior end of female worms was covered densely with minute, cilia-like cuticular spines (Fig. 6).

The eggs of *G. hispidum* had deep pits on their surfaces (Fig. 7). These findings were compatible with SEM observations of adult *G. hispidum* from Shanghai (Koga *et al.*, 1984) and from experimentally infected pigs in Japan (Koga *et al.*, 1984; Kondo *et al.*, 1984).

Discussion

Gnathostoma hispidum is a nematode which lives in the stomach wall of pigs. Adult worms are loosely attached to shallow tumors and are often shed in the feces. This nematode is a relatively common parasite of both domestic and wild pigs in Asia and Europe. It is distributed widely from far eastern to southeastern Asia, but is absent in Japan. In China, few reports concerning the prevalence of adult *G. hispidum* are available. Chen (1936) investigated the helminths of Kwangtung hogs in slaughter houses in Canton Province. He found *G. hispidum* in six of 100 hogs. Wang *et al.* (1976) examined 740 pigs in slaughter houses in Fukien Province. Twenty eight pigs (5.1%) were positive for *G. hispidum*. In the present investigation, sixty seven of 844 pigs (7.9%) were infected in Jiangxi Province which is adjacent to Fukien Province. This prevalence is slightly higher than results of previous surveys of the former two provinces. Jiangxi Province is located in south-central of mainland China. It is surrounded by land, and has some large freshwater lakes in the vicinity of Nanchang where the highest prevalence of *G. hispidum* occurred.

The presence of freshwater lakes in Jiangxi Province provides ideal conditions for this parasite to complete its life cycle. The pigs in this province also roam freely, increasing their potential exposure to *G. hispidum*. Wang *et al.* (1976) demonstrated experimentally that Chinese pigs could be infected by eating fresh water fish harboring the Ad.L_3 and by drinking water containing copepods bearing the E.L_3. Many vertebrates, including loaches may be suitable intermediate hosts in and around fresh water. How pigs become naturally infected has not been determined. And the larvae in the loaches are small in size as E.L_3 in spite of having been reared in vertebrate host is also puzzling. Komaya *et al.* (1945) reported that the Ad.L_3 of *Gnathostoma spinigerum* lived in the viscera and especially livers of fresh water fishes of the Yangtze valley. By contrast, the Ad.L_3 of *G. spinigerum* lives mainly in the muscles of fresh water fish in Japan. The large larvae, as yet unidentified, were found in the viscera of loaches. Koga *et al.* (1985) fed the large larvae to a cat, the final host of *G. spinigerum*, and to two ferrets, a supposing experimental definitive host of *Gnathostoma vietnumicum*. No infection occurred in these animals. It was difficult to carry out satisfactory experimental infections because the number of larvae obtained was small. The host-

parasite relationships of gnathostomes in nature are very complicated. Additional research is needed to determine how the parasites are transmitted under natural conditions. Jiangxi Province seems to be an appropriate place to do those surveys.

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中華人民共和国江西省における剛棘顎口虫の分布

Huang Wen Chang*・Xia Bao Fen*・古賀正崇*・石井洋一*

*中華人民共和国江西医学院寄生虫学教室　*九州大学医学部寄生虫学教室

1980年頃から西日本地域を中心として人体顎口虫症が発生した．これら患者は中華人民共和国，中華民国，大韓民国などから輸入されたと思われるドジョウの生食により感染した．これらドジョウを検索した結果，多数の顎口虫幼虫が得られた．これら幼虫の大部分は小型の幼虫で実験感染の結果，これが剛棘顎口虫*Gnathostoma hispidum* であることがわかった．しかし現在のところ自然界における同虫の生活史は不明であり，またこのドジョウに自然感染している幼虫の発育段階も不明である．この顎口虫の自然界における発育過程を解明するためにはまず成虫の分布調査を行なう必要がある．中国大陸には古くから同虫が分布しているが，今回は中華人民共和国江西省を選び，同省内の高安，奉新，吉水，臨川，上高，豊城，南豊，南昌，新淦，峡江，永修の11地域の調査を実施した．調査方法は同地域屠場で屠殺された豚（中国種）の胃壁を検索した．その結果，奉新地区を除くすべての場所から顎口虫成虫が得られた．最も感染率が高かったのは南昌で豚125頭中17頭が陽性，感染率は13.6%であった．最も低かったのは高安で感染率3.4%であった．全体では豚844頭中67頭に顎口虫の感染が見られ，その感染率は7.9%であった．

海産剛棘顎口虫成虫と同一の形態をしていることが認められた．今回調査した江西省にはかなり広範囲に剛棘顎口虫が分布していることが示唆され，引き続き同省において剛棘顎口虫の発育環追究のための調査が実施されている．