A New Lung Fluke found in Peru, Paragonimus amazonicus sp. n. (Trematoda: Troglotrematidae)

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Introduction

The causative agent of human paragonimiasis in Peru had long been regarded as Paragonimus westermani (Kerbert, 1878) which had been imported by immigrants from the Orient. Recently, however, Ibáñez and Miranda (1967, 68) first found adult lung flukes different from P. westermani in a domestic cat and then in an opossum, Didelphis azarae pernigra from Department of Cajamarca, northern part of Peru, where human paragonimiasis is known to be present. These flukes were named Paragonimus peruvianus by Miyazaki, Ibáñez and Miranda (1969) as a new species. Most recently, Miyazaki and Grados (1972) reported Paragonimus caliensis Little, 1968 in the same area, which was originally found in Cali, Colombia.

On the other hand, Miyazaki, Arellano and Grados (1972) first demonstrated two lung flukes of unknown species, which were obtained in 1970 from a worm cyst in the right lung of a 36-year-old Peruvian male. He was suspected to have been infected in the flat jungle near Aguaytia, Department of Loreto, located in the central part of Peru. Accordingly, the present authors examined some crabs and mammals at the same locality in 1971, but they could not find either adults

or metacercariae of the genus *Paragonimus*. Fortunately, however, they obtained a single metacercaria of the genus from one of 13 crabs, *Pseudothelphusa chilensis*, in a small city, Tingo Maria, Department of Huánuco, which is located about 60 km from Aguaytia and at an altitude of 670 m. Therefore, the authors again visited Tingo Maria in September 1972, and found peculiar lung flukes from two kinds of mammals belonging to Didelphidae. In the present paper the flukes are described as a new species.

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Materials and Methods

Crabs and mammals were trapped along the Huallaga River which runs through Tingo Maria and joins the Amazon River. Twenty-five crabs, Pseudothelphusa chilensis were examined, but none of them were proved to be infected with Paragonimus metacercariae. One of two four-eyed opossums, Philander opossum, captured at Afilador in Tingo Maria harbored two lung flukes, one in a mature worm cyst of the right lung and the other in the pleural cavity, which already died and degenerated. A water opossum, Chironectes minimus (Spanish name: rata de agua), which was trapped at Moyuna in the same city harbored three lung flukes in a young worm cyst of the right lung, but a common opossum, Didelphis marsupialis, was negative for the fluke.

The worms obtained were examined in saline solution for their movement and laying

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eggs, and then fixed with 70% alcohol under pressure. They were brought to Japan, stained with alum-carmine and mounted in balsam, after their cuticle and vitellaria were partly removed over and under the ovary and testes. A piece of the lung containing the mature worm cyst from the infected four-eyed opossum was brought to Japan in 10% formalin.

Measurements were made on four mounted adult specimens and on 60 eggs from the worm cyst preserved in formalin. Structure of the eggshell was demonstrated through the light and the scanning electron microscope. Drawings of the ovary and testes were made by projecting method.

Description of Paragonimus amazonicus sp. n.

Four flukes, except the dead one, moved actively in saline solution, extending their body so strongly that they looked like a kind of nematode. The biggest fluke (holotype) from the four-eyed opossum was fully matured, but other three from the water opossum were young adults containing a small number of immature eggs in their uterus. Two of the three young flukes were slightly damaged in their body.

Holotype (Figs. 1 and 6) extraordinarily elongated, measuring 18.8 by 5.5 mm (ratio 3.4:1). Cuticular spines mostly singly spaced. Oral sucker subterminal, 1.11 mm wide by 0.95 mm long, followed by a small pharynx and a short esophagus. Intestines run posteriorly to the end of body, winding slowly. Ventral sucker 1.09 mm wide by 1.07 mm long, situated anteriorly at 33 % of body length from anterior extremity. Ovary

on the right side of body, 1.41 by 1.26 mm in outline, divided into six lobes, each subdividing into several short lobes, some of them being provided with short processes at Because lobes are mostly short, ovary appears not so delicately branched as a A small receptaculum seminis is recognized. Uterus filled with numerous eggs, situated somewhat bilaterally, extending to the right side of body. Vitellaria densely distributed all over except dorsal and ventral midlines. Testes remarkably small as compared with body size, pressed posteriorly by big uterus. Right testis 2.38 by 0.83 mm in outline, simply divided into six masses, some of them being provided with short Vas efferens from each mass processes. joining to vas deferens clearly recognized. Left testis 2.04 by 0.94 mm in outline, likewise divided into six masses with some processes, but vas efferens partly invisible. Genital pore opens immediately posterior to ventral sucker; no cirrus sac. Excretory bladder reaches near bifurcation of intestines.

Paratype 1 (Figs. 3 and 7, Table 1) smaller but more slender than holotype, ratio of body length to width being 3.6:1. Ventral sucker slightly larger than oral, situated at 31% of body length from anterior extremity. Ovary on the right side, divided more simply than holotype. Uterus contains a few immature eggs only, but slightly extended to ovary side. Testes situated more anteriorly than holotype, seems to be divided into six masses, connection of them being not completely revealed. Other features agree with holotype. Paratypes 2 and 3 (Table 1) almost similar to no. 1, except that ovary of no. 2 lies on the left side of body. Ratio of body

Table 1.	Measurements o	f three	paratypes	of F	amazonicus	รก	n	(in	mm'	١

No.	Body	Oral sucker		Ventral sucker		0	Testis		
		Width	Length	Width	Length	Ovary	Left	Right	
1	11.5×3.2	0.73	0.56	0.85	0.75	0.95×0.65	1.17×0.43	1.39×0.71	
2	10* ×2.7	0.73	0.44	0.77	0.82	1.24×0.51	1.28×0.73	1.26×0.51	
3	11* ×2.8	0.77	0.51	0.75	0.82	0.82×0.60	1.45×0.71	1.00×0.43	

^{*} Approximate length because of partial damage.

length to width 3.7:1 in no. 2 and 3.9:1 in no. 3. Ventral sucker situated at 32% and 30% of body length from anterior extremity in no. 2 and no. 3, respectively.

Eggs (Figs. 2, 4 and 5) laid by holotype yellowish in color and oval in shape; 81 to $102\,\mu$ long by 43 to $55\,\mu$ wide, averaging 90 by $48\,\mu$, when free from pressure. Eggshell thin and uniform in thickness, but with irregular undulation and not rarely with a tail-like process at non-operculated pole.

Host: Philander opossum (four-eyed opossum, type host) and Chironectes minimus (water opossum, rata de agua).

Habitat: Lungs.

Locality: Tingo Maria, District of Rupa-Rupa, Province of Leoncio Prado, Department of Huánuco, Perú.

Type specimens: Department of Parasitology, Faculty of Medicine, Kyushu University, Fukuoka, Japan.

Discussion

Paragonimus amazonicus sp. n. is readily distinguished by its extremely elongated body from all known species of American lung flukes, i.e. P. kellicotti Ward, 1908, P. caliensis Little, 1968, P. mexicanus Miyazaki et Ishii, 1968 and P. peruvianus Miyazaki, Ibáñez et Miranda, 1969. Among Asian species, the present fluke is closest to P. skrjabini Chen, 1959 (syn. P. szechuanensis Chung et Tsao, 1962) in their slender body, singly spaced spines, anterior location of ventral sucker, somewhat delicately branched ovary, and small size of testes. But, they are separated by the character of eggs, because

those of P. skrjabini have no undulation on the shell. The new species is also similar to P. uterobilateralis Voelker et Vogel, 1965 in Africa regarding distribution of their uterus; but in the latter species the body and eggs are apparently smaller and testes are much larger than the former as compared with respective body size. Eggs of P. amazonicus are differentiated from those of Asian and African species ever reported as well as P. kellicotti in North America by the character that eggshells are thin and undulated throughout. This character is the same to the eggs of P. caliensis and P. peruvianus, but it seems that slender form of eggs in the new species may be used for separating them.

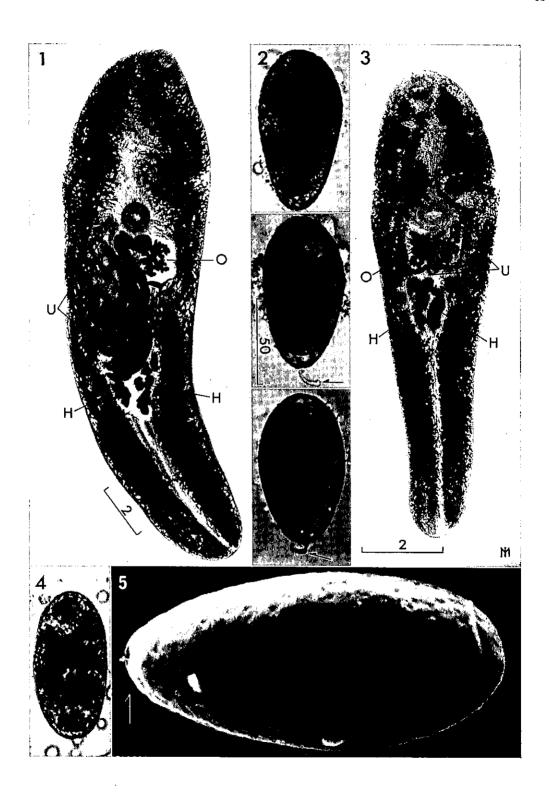
Little (1968) reported lung flukes from wild felines in Colombia without giving their specific names. Some flukes from a margay cat, Felis weidii, seem to be close to P. amazonicus in slender body, singly arranged spines, profusely branched ovary, bilateral location of uterus and eggs resembling P. caliensis, but branching of testes is different from each other. Anyhow, further detailed comparison of them is quite necessary.

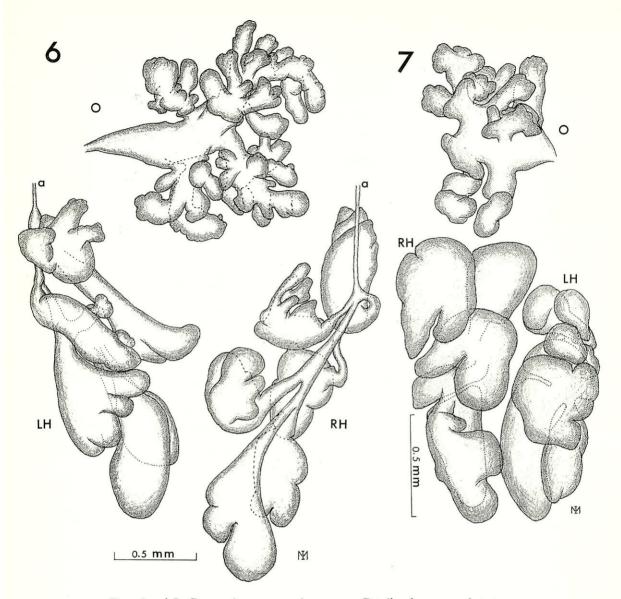
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The single metacerearia, which was found in 1971 by the present authors from *Pseudothelphusa chilensis* in Tingo Maria, seems to belong to *P. amazonicus*, because both were obtained in the same locality. This will be proved in the future by experimental infection. *P. amazonicus* is certainly different from the lung flukes reported in the human lung by Miyazaki, Arellano and Grados (1972), but it probably has some possibility of human infection, because people in Tingo

Explanation of Figures

Figs. 1-5 Paragonimus amazonicus sp. n. 1. Holotype; dorsal view. Cuticle and vitellaria partly removed. Scale in mm. H: testis, O: ovary, U: uterus. 2. Three formalin-fixed eggs laid in the worm cyst by holotype. Same magnification (×500). Scale in micra. Note irregular undulation on the shell. Arrow means taillike process. 3. Paratype 1; ventral view. Scale in mm. Abbreviations same as figure 1. 4. Living egg laid by holotype (×470). Undulation on the shell more clear than figure 2. 5. Scanning electron micrograph of a formalin-fixed egg (×1,200). Undulation of shell clear; taillike process visible (arrow).





Figs. 6 and 7 Paragonimus amazonicus sp. n. Details of ovary and testes, anterior end at top. O: ovary, LH: left testis, RH: right testis, a: vas deferens. 6. Holotype; dorsal view. 7. Paratype 1; ventral view.

Maria occasionally eat the same kind of crab, which harbored *Paragonimus* metacercaria of unknown species mentioned above. In so far as the authors are aware, the water opossum is a new host for the lung fluke.

Summary

A peculiar lung fluke, Paragonimus amazonicus sp. n. was described from the lungs of two of four mammals, a four-eyed opossum, Philander opossum and a water opossum, -Chironectes minimus in Tingo Maria, Department of Huánuco. For identification of the new species the following points are important: 1) body is extraordinarily elon-.gated, 2) cuticular spines are mostly singly .spaced, 3) oral sucker is about the same size with the ventral, which is located very anteriorly, 4) ovary is somewhat delicately branched, 5) testes are divided into six masses and slightly larger than ovary, but very small as compared with body size, 6) uterus is located somewhat bilaterally, and 7) eggs are slender and eggshells are thin The crab host is presumed and undulated. to be Pseudothelphusa chilensis, but the metacercaria is unknown at present. There is some possibility of human infection with the new species.

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ペルーで発見されたアマゾン肺吸虫(新称)

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ペルー中部, Tingo Maria で、1971年にチリーサワガニから未知の肺吸虫幼虫をみつけた。 そこで72年に再び同地を訪れた結果、2種のフクロネズミから、新しい肺吸虫をみつけた。 体が非常に細長い点で、既知のアメリカ産4種とは容易に区別できる。アジア産の中では、中国原産のスクリアビン肺吸虫に最も近いが卵が明

らかに異なる. 結局, どの既知種にも一致しないので, 新種として報告した. これはペルー産肺吸虫の第3種と なる. メタセルカリアは確認していないが, 上述のカニ が宿主であることは, まず間違いない. これを住民がと きどきたべるので, 人体感染もありうる.