

Comparative Studies on Two Forms of *Paragonimus* Parasitic in *Eriocheir japonicus* on Cheju Island, Korea (Preliminary Report)

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During their investigations on the infection of *Paragonimus westermani* (Kerbert, 1878) in crustacean hosts in Korea, Kim and Lee noticed an apparent difference in size among the cysts of the metacercariae collected from crabs, *Eriocheir japonicus* which had been collected on Cheju Island. They divided the metacercariae into three groups tentatively, based on sizes; large-form, middle-form and small-form. As an initial approach to distinguish any differences in morphology in the adult stage and in eggs of the respective forms of the metacercariae, those of the large- and the small-form were fed to two dogs separately. After 88 days of infection the dogs were sacrificed and examined. They found that the adult worms and eggs collected from the dog fed with small-form metacercariae were both smaller with a difference in shape of eggs compared with those collected from the dog fed with the large-form metacercariae.

On the occasion Miyazaki visited Korea in May 1970, he was consulted with these interesting results and some of the specimens of adults and eggs were brought to Japan for reexamination. Miyazaki and Hashiguchi investigated them again in detail, and confirmed the above-mentioned size difference

between the two forms. In this paper the authors wish to report preliminary results.

Materials and Methods

The metacercariae collected from a number of *E. japonicus* by the method of grinding and digestion were divided into three groups based on the maximum diameter of their inner cysts. The large-form measured from 389 to 450, with an average of 415 μ , and the small-form from 259 to 300, with an average of 283 μ . The metacercariae measuring between these two groups were designated as the middle-form. One hundred larvae of the large- and the small-form each were orally given to two adult dogs separately. The dogs were sacrificed 88 days after infection and the mature worms were collected from the worm cysts of the lungs of each dog. The majority of the adult worms obtained were lyophilized for antigen preparation and the remaining part was flattened and fixed in FAAG fixative and preserved in 3% glycerin alcohol. Some of the fixed specimens were stained with carmine and mounted in balsam for morphological investigation. Fresh eggs of both forms were collected from worm cysts in the lungs of the respective dog and preserved separately in 10% formalin solution.

In this paper, the terms of "L-form" and "S-form" used in adult and egg stages mean that they originated from the large-

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and the small-form of metacercariae, respectively. Adult specimens reexamined in Japan were two mounted and four formalin-preserved L-forms and seven mounted S-forms. One hundred eggs of each form were measured free from pressure and carefully examined for the shape of whole body and the thickness of eggshell at non-operculated end. Furthermore, 100 uterine eggs removed from each of four preserved adults of L-form were measured without pressure. The length of each ten eggs deposited in the terminal part of uterus was also measured in the mounted adult specimens of two L-forms and seven S-forms.

Results

As shown in Fig. 1, size difference of adult body was clearly demonstrated between L- and S-form in living condition. The same difference was likewise recognized in mounted specimens of both forms as shown in Table 1. But, adults of both forms were not differentiated from each other by common morphological criteria such as in ovary, testes, suckers and cuticular spines.

Table 1 Comparison of the two forms in mounted specimens

Form	Adult no.	Body size (in mm)	Aver. length of 10 eggs in uterus (in micra)
L	1	12.5×5.7	89.0
	2	11.7×6.0	94.5
S	1	8.6×4.0	65.0
	2	8.8×4.3	68.3
	3	8.3×4.5	69.8
	4	7.6×3.5	64.8
	5	8.3×4.4	67.8
	6	8.4×4.3	73.3
	7	8.6×4.6	69.8

On the other hand, 100 eggs of L-form laid in worm cysts measured 75.0 to 105.0 (91.70 ± 4.40) by 37.5 to 57.5 (47.70 ± 3.14) μ (mean \pm standard deviation), and 100 eggs of S-form measured 60.0 to 95.0 (70.18 ± 5.39) by 35.0 to 52.5 (43.68 ± 3.47) μ . The differ-

ence of both length and width between the two forms was statistically significant ($P < 0.001$). In other words, most eggs of L-form were more elongated than those of S-form. Out of 100 eggs of L-form examined, the thickening of eggshell at non-operculated end was remarkable in 28, moderate in 57, slight in 12 and negative in three; on the contrary, in case of S-form the thickening was remarkable in five, moderate in 25, slight in 48 and negative in 22 out of 100 eggs examined. These facts showed that the thickening of shell at non-operculated end was more common and more conspicuous in L-form than the other (Figs. 2-5). Each 100 eggs removed from terminal part of the uterus of four adult L-forms, which were preserved in 10% formalin, averaged 95.7 by 47.0, 93.8 by 49.2, 89.6 by 48.6 and 93.4 by 48.4 μ , respectively. These eggs showed almost similar size and shape to those of L-form collected from worm cysts of the lungs mentioned above.

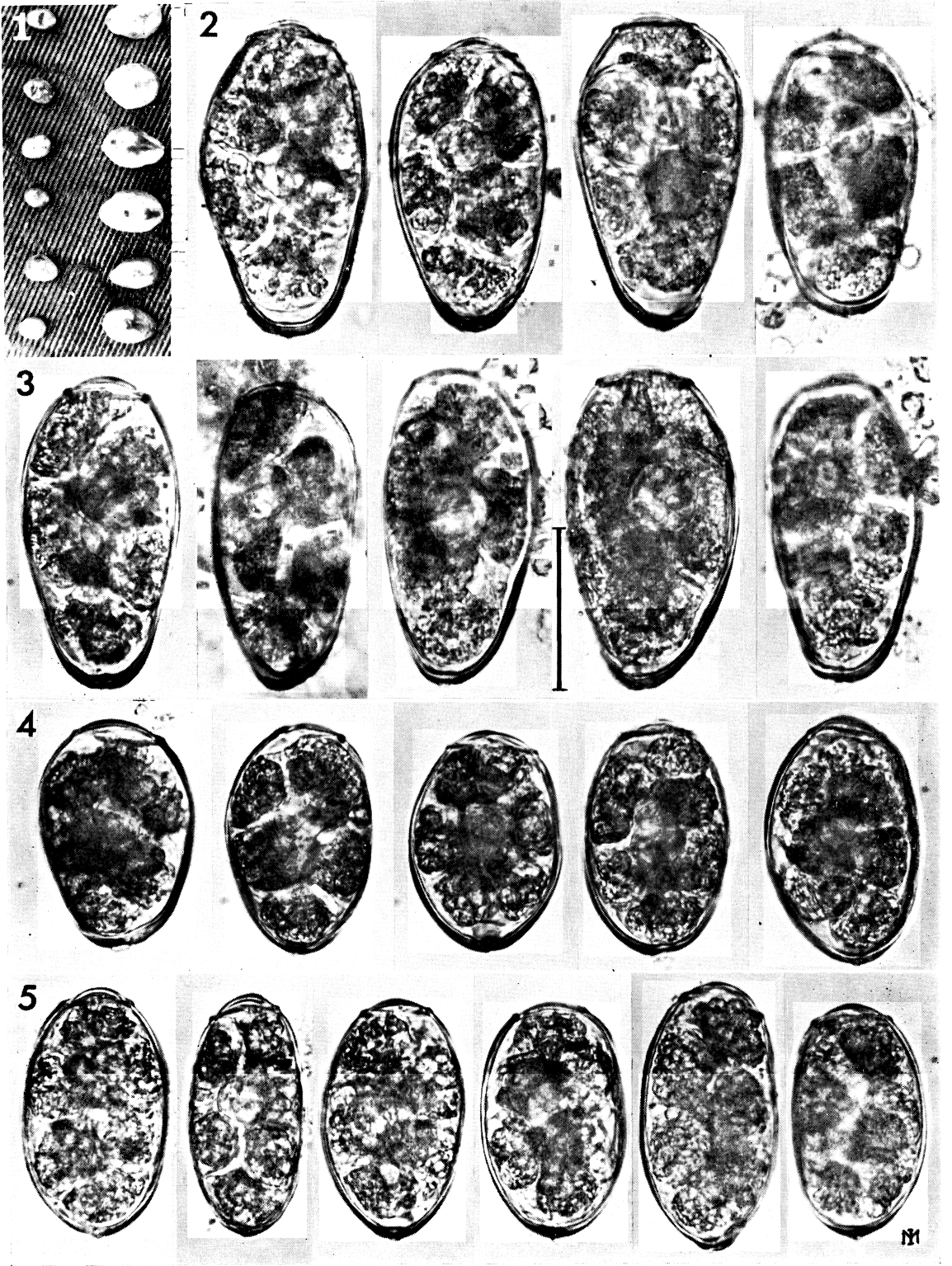
In addition, the length of each ten eggs were measured in the terminal part of the uterus of mounted specimens, and it demonstrated clear difference between the two forms, as shown in Table 1.

Discussion

Paragonimus metacercariae parasitic in *E. japonicus* have long been believed to belong exclusively to *P. westermani*, and any other species of the genus have never been reported from this crab throughout Asia. The present study, however, suggests that in the future more than one species or subspecies of *Paragonimus* may be found from this crab.

It is certainly true that the L-form in this paper belongs to the genuine *P. westermani*, but with regard to the S-form, there is hesitation in identifying it as *P. westermani*. The authors are inclined to think that the S-form has some possibility to be a different species or subspecies. In order to resolve this question, it is quite necessary to investigate more materials particularly metacercariae in detail.

In China, Chung and Tsao (1962) reported briefly on their "*Paragonimus westermani*



(Szechuan variety)", which was found in two kinds of crabs, *Potamon denticulatus* and *P. yaanensis*. According to them, metacercariae of this variety averaged 280 by 262 μ in diameter, and its eggs measured 74.4 by 46.4 μ on the average. These figures are close to those given in this paper for the S-form. Although Chen (1963) was contradictory to "Szechuan variety", it needs further detailed investigation for resolution of this question.

Summary

On Cheju Island, Korea, *Paragonimus* metacercariae parasitic in *Eriocheir japonicus* were divided into three forms according to the diameter of their inner cysts. The

large-form and the small-form were fed to two dogs separately, and both adults and eggs originated from the small-form were obviously smaller than those originated from the large-form. It is likely that there is other species or subspecies than the genuine *Paragonimus westermani* on the Island.

Literatures Cited

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韓国濟州島のモクズガニに寄生する肺吸虫2形の比較 (予報)

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韓国産のカニについて、ウェステルマン肺吸虫をしらべていた金と李は、濟州島で集めたモクズガニに寄生するメタセルカリアの大きさに、かなりの変異があることに気づいた。そこで、これらを大中小の3群にわけ、そのなかの大形と小形を100ずつ別々のイヌにたえて、88日後に剖検したところ、大形から発育した成虫および卵は、小形からのものに比べて、明らかに大きいことに気づいた。

1970年の5月に訪韓した宮崎は、この問題について意見をきかれたので、その材料をもちかえり、橋口とともに、くわしい再検討をつづけた。その結果、成虫の大きさは明らかにちがうが、従来の鑑別点には差がみられ

ないこと、また、卵は大きさに明らかな差があるばかりでなく、形も異なり、とくに無蓋端にも差のあることをみとめた。そして、大形の方は、容易に、ウ肺吸虫と同定できたが、小形は同定をちゆうちよせざるをえなかつた。中国大陸で、Chung and Tsao (1962) が報告した *Paragonimus westermani* (Szechuan variety) に対して、Chen (1963) は否定的であるが、小形はこれに近いように思われる。この問題を解明するには、なお多くの材料を必要とし、とくに、生きたメタセルカリアをくわしく比較しなければならないので、ここには、予報として、上記の事実を記録するにとどめた。

Explanation of Figures

Fig. 1 Living adults of L-form (right) and S-form (left). Unit of scale: 1 mm. Specimens were placed on a dry paper background. (Photographed by Kim)

Figs. 2-5 Eggs of L-form (2 and 3) and S-form (4 and 5) under the same magnification. Scale: 50 μ . Removed from the worm cysts and preserved in 10% formalin. (Photographed by Miyazaki)