# Studies on Intestinal Trematodes in Korea

# III. Natural Human Infections of *Pygidiopsis summa* and *Heterophyes heterophyes nocens*

# Byong-Seol Seo, Sung-Tae Hong and Jong-Yil Chai

Department of Parasitology and Institute of Endemic Diseases College of Medicine, Seoul National University

#### INTRODUCTION

The small intestinal flukes of the family Heterophyidae Ohdner, 1914 are classified into 22 genera (Yamaguti, 1958). Total 8 genera among them, i.e., Heterophyes, Heterophyopsis, Metagonimus, Stellantchasmus, Centrocestus, Haplorchis, Pygidiopsis and Procerovum had been reported human parasitic (Komiya et Suzuki, 1966). However, human infections other than these genera are supposed to be possible also, because heterophyid fluke infections are one of zoonoses in nature.

When they infect human intestines, severe enteritis may occur. And the symptoms caused by them are closely related with the number of infected worms. Really, their infections tend to be passed unnoticed when the worm burden is low. Furthermore, *Heterophyes*, *Stellantchasmus* and *Haplorchis* were reported to cause fatal extraintestinal heterophyidiasis by spreading eggs into the blood stream (Africa et al., 1940).

Among the heterophyid flukes in Korea, *Metagonimus yokogawai* is the only species that was proved to be prevalent in inhabitants along several rivers and streams, and its medical importance has been studied sufficiently until now. However, even a human case of heterophyid flukes other than *M. yokogawai* has not been recorded yet. Only the distribution of some flukes was recorded by detecting metacercariae

from piscine hosts. Heterophyes was reported by Seo et al. (1980), Stellantchasmus was by Seo et al. (1979), Centrocestus was by Choi et al. (1964) and Pygidiopsis was by Chun (1963).

The authors found small heterophyid eggs which were very similar to those of Clonorchis sinensis but apparently smaller in size, from 8 cases during a stool examination of inhabitants in Okku area, North Cholla Do (Fig. 1) in 1979. And all the cases were proved to be infected with Pygidiopsis summa by obtaining adult worms after treatment with bithionol. And from the most heavily infected case, a few worms of Heterophyes and Metagonimus were collected concomitantly. Therefore, this is the first record on human infection cases of Pygidiopsis and Heterophyes in Korea.

#### MATERIALS AND METHODS

Detection of the eggs was made by cellophane thick smear and formalin-ether concentration techniques, and Stoll's egg counting method was also applied.

For treatment on the egg positive cases and adult worm collection, bithionol was given in dose of 40 mg/kg body weight being divided into two doses with one hour interval, after premedication with 30gm of magnesium sulfate. Two hours after the administration of bithionol, magnesium sulfate 30gm was used for three times successively with one hour intervals. All the

watery diarrheal stool were collected from each case and the worms discharged from them were picked out under a dissecting microscope.

The collected worms were washed in saline solution and their numbers were counted in each case. Some of the worms were observed under a cover slip in saline solution and then fixed in 10% formalin. Well-fixed worms were stained with Semichon's acetocarmine and observed.

#### RESULTS AND WORM DESCRIPTION

1. On the human infection cases of Pygidiopsis: All the cases were personnel of a salt farm company in Okku Gun, North Cholla Do (Fig. 1) and aged 32~52 years. They used to eat the raw flesh of brackish water fishes preferably, which were caught in a sea-water reservoir in the salt farm at the mouth of the Mangyong River. They had not any significant symptoms or signs caused by this infection. Clinical laboratory examination was taken only

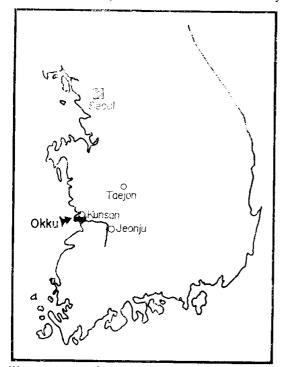


Fig. 1. A map showing Okku area, North Cholla Do at the mouth of Mangyong River.

**Table 1.** Results of egg counting and worm collection of heterophyid flukes

Case	Age	EPG of heterop-		ber of sollected	l	Adverse effects
		hyid egg	opsis_	Hetero phyes	- Metag onimus	of drugs
1	52/M	1, 200	4, 045	3	7	
2	48/M	200	982	0	0	
3	$33/\mathbf{F}$	100	674	0	0	
4	$49/\mathbf{M}$	0	183	0	0	
5	42/M	500	371	0	0	Dizziness
6	51/M	100	68	0	0	
7	46/M	0	395	0	0	Epigastric
8	35/ <b>M</b>	400	1,098	0	0	pain

for the most heavy burden case (Case 1). And the eosinophil count was 7% and the results of urinalysis, CBC and blood chemistry were all in their normal ranges.

Table 1 shows the results of egg counting and worm collection from the 8 cases. The EPG count was in the range of  $0\sim1,200$  and the collected worms were counted from 68 to 4,045 in a case. The values of EPG were relatively low compared with the numbers of collected worms. Adult worms of Heterophyes and Metagonimus were also recovered 3 and 7 in number respectively from the Case 1.

The adverse effects due to the administration of bithionol and magnesium sulfate were observed in two cases. Mild degree dizziness and epigastric pain were noted in Case 5 and 7 respectively (Table 1).

2. On the worm identified as *Pygidiopsis* summa: A total of 10 worms was measured after staining and the morphological descriptions are as follows.

Body dorsoventrally flat, tapering anteriorly, and globular and bluntly ending posteriorly. Lateral margins of posterior half of body curved ventralwards and ventrally scaphoid when not pressed (Fig. 2). Fine spines covered the surface of body except for the posterior one third. Body size  $0.412 \sim 0.738$ mm long and  $0.279 \sim 0.479$ mm

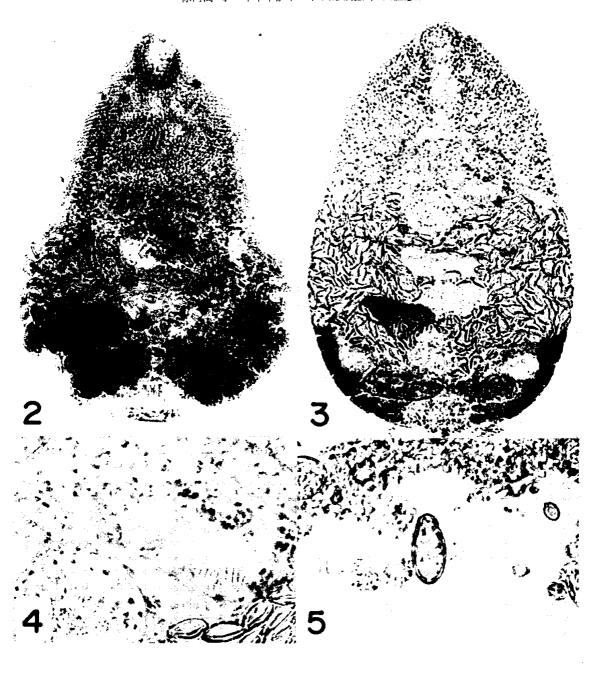


Fig. 2. Adult Pygidiopsis summa, unpressed and acetocarmine stained,  $\times$  180.

Fig. 5. Egg of Pygidiopsis summa,  $\times 400$ .

Fig. 3. Adult Pygidiopsis summa, pressed and acetocarmine stained, ×200.

Fig. 4. High power view of the genital apparatus showing gonotyls,  $\times 400$ .

wide. Oral sucker subterminal, measured 0.055~ 0.063×0.040~0.055mm. Prepharynx present, 0.004~0.035mm long. Pharynx, well developed and muscular, 0.028~0.047mm long and  $0.030\sim0.044$ mm wide. Esophagus  $0.040\sim$ 0.095mm long and bifurcated into two intestinal ceca near the anterior one third line of body. The ceca, tubular to lateralwards and posteriorwards along the lateral body lines and ending at the anterior margins of testes straightly overlapping the vitelline follicles. Excretory vesicle not so easily seen but excretory pore visible at the midline of posterior body end. Ventral sucker, at the middle portion of body, measured 0.051~0.079×0.047~0.063mm. Neighboring the ventral sucker, genital apparatus present left anterolaterally, 0.063~0.095mm long and 0.032~0.047mm wide. Genital apparatus crescent shaped and included male genital accessories and genital openings though not so easily observable. Two groups of small spinelike gonotyls (Fig. 4) present at the posterior portion of genital apparatus, one consisted of 5 and the other 4, however, not so commonly seen in all specimens. Seminal vesicle connected to genital apparatus,  $0.071 \sim 0.134 \times 0.047 \sim 0.071$ 

mm, and bilobed by a constriction and sausage shaped. Seminal receptacle, globular and 0.079~ 0.126×0.047~0.087mm, locating just anteriorly to testes at midline. Small thread-like sperms visible showing whirl-pattern in seminal vesicle and receptacle. Right and left testes, transversely oval and symmetrical at the posterior body end, and measured  $0.070{\sim}0.126{\times}$ 0.047~0.060mm. Ovary, globular, round to elliptical, located anterior to right testis, measured 0.060~0.111×0.040~0.071mm. Uterus, filled with eggs, coiled between ventral sucker and testes, and reached at the lateral margins of body. Vitelline glands formed 6~8 follicles in a row in each outside of ovariotesticular zones (Fig. 3). The measurements are summarized in Table 2. From above descriptions, these worms were identified as Pygidiopsis summa Onji et Nishio, 1916.

A total 25 eggs (Fig. 5) of *Pygidiopsis* measured  $0.018\sim0.022\times0.011\sim0.013$ mm (mean  $0.020\times0.012$ mm). The eggs were ovoid, operculated, and thick shelled, very similar to those of *C. sinensis*, but smaller in size and some eggs were rather stout. Shouldering and shell wrinkling were not so distinctive. Shell color was

Table 2. Measurements of adult Pygidiopsis summa

Organ	Range(mm)	Mean(mm)
Length	0.412~0.738	0. 627
Width	0.279~0.479	0. 366
Oral sucker	$0.055\sim0.063\times0.040\sim0.055$	0. 060 × 0. 050
Prepharynx	$0.004\sim 0.035$	0. 015
Pharynx	0. 028~0. 047×0. 030~0. 044	$0.038 \times 0.036$
Esophagus	0.040~0.095	0.056
Ventral sucker	$0.051 \sim 0.079 \times 0.047 \sim 0.063$	$0.065 \times 0.055$
Genital apparatus	0.063~0.095×0.032~0.047	0. 075×0. 038
Ovary	$0.060\sim 0.111\times 0.040\sim 0.071$	$0.081 \times 0.057$
Γestes	$0.070\sim 0.126\times 0.047\sim 0.060$	$0.096 \times 0.051$
Seminal vesicle	$0.071 \sim 0.134 \times 0.047 \sim 0.071$	$0.095 \times 0.057$
Seminal receptacle	$0.079 \sim 0.126 \times 0.047 \sim 0.087$	$0.093 \times 0.037$ $0.101 \times 0.069$
Vitelline follicle	6∼8 in each side	0.101 ^ 0.009

Table 3. Measurements of Heterophyes heterophyes nocens

Organ	Range(mm)	Mean(mm)	
Length	0. 808~0. 942	0. 875	
Width	0.385~0.519	0.452	
Oral sucker	$0.063\sim0.066\times0.070\times0.072$	$0.065 \sim 0.071$	
Prepharynx	0.016~0.024	0.020	
Pharynx	$0.041 \sim 0.047 \times 0.040 \times 0.052$	$0.044 \times 0.046$	
Esophagus	0.066~0.079	0.073	
Ventral sucker	$0.165\sim 0.167\times 0.119\sim 0.166$	$0.166 \times 0.143$	
Genital sucker	$0.133\sim 0.166\times 0.100\sim 0.106$	$0.150 \times 0.103$	
Testis	0.100~0.106×0.073~0.100	$0.103 \times 0.087$	
Ovary	0.080~0.133×0.066~0.093	$0.107 \times 0.080$	
Seminal vesicle	$0.100\sim0.119\times0.066\sim0.073$	$0.110 \times 0.070$	
Seminal receptacle	$0.199 \times 0.139$	$0.199 \times 0.139$	
Vitelline follicle	5 in each side		

yellowish brown and each egg contained mature miracidium.

3. On the worm, Heterophyes heterophyes nocens: The worms collected from the first case measured 0.808~0.942mm long and 0.385~0.519mm wide. Body dorsoventrally flat and ovoid in shape and the morphological characteristics and measurements were almost same as those of Seo et al. (1980). The measurements are summarized in Table 3. The number of rodlets around genital sucker was in range of 54~57, and the worms were identified as Heterophyes heterophyes nocens. The eggs of Heterophyes were not found from the Case 1.

#### DISCUSSION

The genus *Pygidiopsis* was firstly described by Looss (1907). The type species was *P.genata* from the intestine of a pelican in Egypt. The morphological characteristics of this genus were globular and scaphoid shape of the hindbody, crescent shaped genital apparatus located left anterolaterally adjacent to ventral sucker, bilobed seminal vesicle and vitelline follicles in lateral fields of the ovariotesticular portion.

The worms collected from the 8 cases in this study are well corresponding to the original description of genus *Pygidiopsis*.

For the species of Pygidiopsis other than P. genata, P. summa was reported in Japan by Onji et Nishio, 1916; P. pindoramensis was in Brazil by Travassos, 1929; P. plana was in North America by Price, 1934; P. marivillai was in the Philippines by Refuerzo et Garcia, 1937; P. phalacrocoracis in Japan by Yamaguti, 1939 and P. piclaumoreli was in Senegal by Dollfus et Capron, 1958. These speciations based mainly on the measurements and the relative size differences of body or organs. And the geography of the areas where the above worms were collected seems to have been another factor for the speciation.

Among them, the whole life cycle of *P. summa* was outlined by Ochi (1931) in Japan. He found ophthalmopleurolophocercous cercariae different from those of *Heterophyes* from a kind of brackish water snail, *Tympanotonus microptera*. Afterwards, *Mugil cephalus*, *Acanthogobius flavimanus* and *Liza menada* had been reported as the second intermediate hosts of this species in Japan. The final hosts were found as dogs

(Asada, 1927), cats, *Milvus migrans lineatus* and *Nyctocorax nyctocorax* (Komiya, 1965). As for the human cases of *P. summa*, Takahashi (1929) reported firstly by detecting egg, and Asada (1952) detected 5 carriers and Yokogawa et al. (1965) added 4 cases by obtaining adult worms with kamala treatment.

When the data of measurements only are considered, the pesent worm well corresponded to *P. summa* which was proposed as a distinctive species because of the larger body size and larger ventral sucker than oral one whereas in *P. genata* ventral sucker was slightly smaller than the oral one.

The differences in morphology of present worm from the above two species are the ceca termination and presence of gonotyls. The ceca of present worm terminate straightly and adjacently to the lateral body lines overlapping the vitelline follicles near testes. However, the ceca of the two species were described as terminating bended dorsomedially at pretesticular level. The spine-shaped gonotyls, grouped in genital apparatus, is another finding that has never been recorded before, though not evenly observed in every worm.

Though there were some morphological discrepancies, the present worms were identified as *P.summa*, putting aside the validity of the previous speciations on the genus *Pygidiopsis*. The reasons of our identification are 1) Japanese parasitologists described the whole life cycle of this species, that made *P. summa* valid, 2) geographical adjacency between Korea and Japan.

A few worms of genus Heterophyes collected in present study were identified H. heterophyes nocens. See et al. (1980) summarized the debates on the speciation of the genus Heterophyes and concluded the worms in Korea they obtained were H. heterophyes nocens. The number of rodlets around genital sucker was said to be a

criterion for the subspeciation of H. heterophyes. The number was  $50\sim60$  in H. heterophyes nocens and  $74\sim87$  in H. heterophyes. The number of present specimen is  $54\sim57$  and thus falls in the subspecies H. heterophyes nocens.

According to Seo et al. (1981), the mullets captured in the reservoir of the salt farm in Okku area harboured many metacercariae, of *P. summa* in their gills. By these findings pygidiopsiasis in Okku area can be prevented by simple evisceration of mullets especially including gills. However, the knowledge on the life cycle of this species in that area can provide more effective methods in control pygidiopsiasis.

#### SUMMARY

During stool examination of the inhabitants in Okku area, North Cholla Do, in December, 1979, 8 cases were found to pass out small heterophyid eggs (measuring  $0.020 \times 0.012$ mm). And follow-up treatment on the egg positive cases with bithionol expelled *Pygidiopsis* and *Heterophyes* spp.

The results of the worm collection and identification are as follows.

- 1. Heterophyid egg positive cases were 8 in number, and the range of EPG count was  $0\sim$  1,200. And the number of collected worms of *Pygidiopsis* was  $68\sim4,045$  per case. Among them, a case was found infected with *Heterophyes* worms 3 in number, concomitantly. These cases did not show any noticeable clinical symptom or sign due to the infections, even the heavy infection case either.
- 2. By their morphological characteristics and measurements, the worm *Pygidiopsis* was identified as *P. summa* Onji et Nishio, 1916. However, there were two distinctive morphologic features in our worms. The ceca terminated straightly in lateral zones of testes overlapping vitelline follicles. And the spine-like gonotyls

were found in the genital apparatus in some worms.

3. The rodlets around genital sucker of present *Heterophyes* were 54~57 in number and identified as *H. heterophyes nocens*.

This report is the first description on the natural human infection of *Pygidiopsis summa* and *Heterophyes heterophyes nocens* in Korea.

### =국 문 초 록=

## 한국의 장흡충류에 관한 연구

# ■. Pygidiopis summa와 Heterophyes heterophyes nocens의 인체감염례

서울대학교 의과대학 기생충학교실 및 풍토병연구소 徐 丙 髙·洪 性 台·萘 鍾 一

전북 옥구군의 주민 검변중에 간흡충란과 비슷한 모양이면서 크기가 더 작은 異形吸蟲類의 충란을 발견하고, 그 감염자에게 비치오놀 40mg/kg를 투여하였다. 그 결과 8例에서 Pygidiopsis 성충을 얻어 우리나라에서는 처음으로 인체감염례를 발견하였다. 그중 1例는 異形吸蟲과의 혼합감염이었다. 그 결과를 요약하면 다음과 같다.

- 1. 8例의 충란양성자에서 EPG는 0~1,200의 법위에 있었고, *Pygidiopsis* 성충은 68~4,045가 각 例에서 수집되었다. 이들 중에 한 例에서는 3마리의 異形吸蟲을 수집할 수 있었다. 이들에게서 별다른 임상적인 증후나 중상은 보이지 않았다.
- 2. 수집된 Pygidiopsis는 형태와 계측치에 의해서 P. summa로 동정하였고, 아직 기록되지 않은 小棘 (gonotyls)을 生殖帶에서 관찰하였다. 또 腸管의 끝이 錄體 中央部로 굽혀지지 않은 채 곧게 되어 있는 점은 이정의 기록과는 다른 소견이었다.
- 3. 異形吸蟲은 生殖盤이 뚜렷하고, 생식반 주위의 Rodlets의 수가 54~57의 범위에 있어 H. heterophyes nocens로 동정하였다.

이상과 같이 Pygidiopsis와 異形吸蟲의 인체감염례를 확인하였으나 임상적으로는 별 특징이 없었다. 이 이외의 異形吸蟲類의 감염도, 소형 흡충란의 감별에 보다 주의를 한다면 더 찾아낼 수 있을 것으로 확신하였다.

#### **ACKNOWLEDGEMENT**

The authors would like to express their great thanks to Mr. Keun-Sik Kang for his sincere and willingly cooperation in detecting the heterophyid egg positive cases.

#### REFERENCES

- Asada, J.: On a new trematode found in the dog of the vicinity of Tokyo with reference to the distribution of trematodes in dogs. Tokyo Iji Shinshi, (2564): 926-930, 1927 (in Japanese).
- Asada, J., Kaji, F. and Ochi, K.: Therapeutic study on the heterophyid trematodes of man. Nippon Kiseichu Gakkai, 21: 67-68, 1952 (in Japanese).
- Choi, D.W., Shin, D.S. and Lee, S.W.: Studies on the larval trematodes from brackish water fishes I. Observation of Centrocestus asadai Mishima, 1959. Korean J. Parasit., 2(1): 14-19, 1964(in Korean).
- Chun, S.K.: On some trematodes whose intermediate hosts are brackish water fishes (II). The life history of Pygidiopsis summus the intermediate host of which is Mugil cephalus. Bull. Busan Fish. Coll., 5(1): 1-5, 1963 (in Korean).
- Dollfus, R.P. et Capron, A.: Un Pygidiopsis de sterna, de la côte du Sénégal. Bull. de l'Institut Français d'Afrique Noire Série A: Sciences Naturelles, 20 (2): 306-310, 1958 (cited from Helminth. Abstract, vol. 27, 1958).
- Komiya, Y.: Metacercariae in Japan and adjacent territories. Prog. Med. Parasit. in Japan, 2: 178-180, 1965.
- Komiya, Y. and Suzuki, N.: The metacercariae of trematodes belonging to the family Heterophyidae from Japan and adjacent countries. Japanese J. Parasit., 15(3):208-214, 1966(in Japanese).
- Looss, A.: Notizen zur Helminthologie Aegyptens. VII. Ueber einige neue Trematoden der aegyptischen Fauna. Centralbl. Bakt., 43: 478-490, 1907.
- Ochi, S.: Studies on the trematodes whose intermediate hosts are brackish water fishes-on the life cycle of Pygidiopsis summus. Tokyo Iji Shinshi, (2712): 346-353, 1931 (in Japanese).

- Price, E.W.: New trematode parasites of birds. Smithson Misc. Coll., 91(6): 1-6, 1934 (cited from Systema Helminthum vol. 1).
- Refuerzo, P.G. et Garcia, E.Y.: Pygidiopsis marivillai, a new heterophyid trematode from the Philippines. Philippines J. Science, 64(4): 359-363, 1937 (cited from Helminth. Abstract, vol. 6, 1937).
- Seo, B.S., Cho, S.Y., Chai, J.Y. and Hong, S.T.: Studies on intestinal trematodes in Korea (II) Identification of the metacercariae of Heterophyes sp. and Stellantchasmus sp. from mullets of Youngsan River and Geoje Do. Korean J. Parasit., 17(2): 165-166, 1976 (Korean abstracts).
- Seo, B.S., Cho, S.Y., Chai, J.Y. and Hong, S.T.: Studies on intestinal trematodes in Korea II. Identification of the metacercariae of Heterophyes heterophyes nocens in mullets of three southern coastal areas. Seoul J. Med., 21(1): 30-38, 1980.
- Seo, B.S., Hong, S.T., Chai, J.Y. and Cho, S.Y.: Studies on intestinal trematodes in Korea IV. Geographical distribution of Pygidiopsis and Heterophyes metacercariae. Seoul J. Med., 22(2): 236-242,

- 1981.
- Takahashi, S.: On the eggs of Stellantchasmus falcatus and Pygidiopsis summus found in human stools.

  Okayama Igakkai Zasshi, 41(7): 1502-1513, 1929

  (in Japanese).
- Travassos, L.: Sur une nouvelle espece du genre Pygidiopsis, P. pindoramensis n. sp. Compt. Rend. Soc. Biol., 100(11): 956-957, 1929.
- Witenberg, T.: Studies on the trematode-family Heterophyidae. Am. Trop. Med. Parasit., 23: 131-268, 1929.
- Yamaguti, S.: Studies on the helminth fauna of Japan.

  Part 25. Trematodes of birds, IV. Japanese J.

  Zoology, 8(2): 129-210, 1939 (in Japanese).
- Yamaguti, S.: Systema Helminthum. vol. I. Digenetic trematodes of vertebrates. Interscience Publishers, New York, 1958.
- Yokogawa, M., Sano, M., Itabashi, T. and Kachi, S.: Studies on the intestinal flukes II. Epidemiological studies on heterophyid trematodes of man in Chiba Prefecture. Japanese J. Parasit., 14(6): 577-585, 1965 (in Japanese).