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Gorgoderid trematodes (Digenea: Gorgoderidae) from the urinary bladder of frogs in Korea

Ki-Hong KIM*[1], Kyoung-Hwan JOO[2] and Han-Jong RIM[2]

Department of Fish Pathology[1], College of Fisheries Sciences, National Fisheries University of Pusan, Pusan 608-737 and Department of Parasitology and Institute for Tropical Endemic Diseases[2], College of Medicine, Korea University, Seoul 136-705, Korea

Abstract: Two species of the family Gorgoderidae (Trematoda: Digenea), *Gorgodera japonica* Yamaguti, 1936 and *Gorgoderina bombinae* Yu & Lee, 1983 were collected from the urinary bladder of frogs captured from various localities in Korea. The morphology of each species is described and illustrated. *Gorgodera japonica* differs from *G. cygnoides* by having deeply branched vitellaria. *Gorgoderina bombinae* is considered a valid species.
Key words: Frog, classification, morphology, *Gorgodera japonica*, *Gorgoderina bombinae*

INTRODUCTION

The family Gorgoderidae Looss, 1901, is a group within the Digenea which includes species usually parasitic in the ureters or urinary bladders of fishes, amphibians and reptiles. The genera of Gorgoderidae found in amphibians are *Gorgodera* Looss, 1899, *Phyllodistomum* Braun, 1899, *Gorgoderina* Looss, 1902 and *Progorgodera* Brooks & Buckner, 1976 (Prudhoe & Bray, 1982).

Two species of gorgoderids are known to inhabit Korean frogs. *Gorgodera japonica* Yamaguti, 1936 parasitizes *Rana nigromaculata* (Song *et al.*, 1965; Lee *et al.*, 1976; Yu & Lee, 1983) and *Gorgoderina bombinae* Yu & Lee, 1983 parasitizes *Bombina orientalis*.

In this study, we redescribed their morphological characters and discussed their systematic status.

MATERIALS AND METHODS

From February 1989 to August 1994, *Bufo bufo gargarizans* (33 individuals), *Bufo stejnegeri* (5), *Hyla arborea japonica* (73), *Kaloula borealis* (6), *Bombina orientalis* (229), *Rana nigromaculata* (293), *Rana plancyi chosenica* (10), *Rana dybowskii* (120), *Rana rugosa* (72), and *Rana catesbeiana* (9) were collected from various locations in Korea and examined for parasites.

Rana nigromaculata and *Bombina orientalis* were infected with gorgoderid trematodes in their urinary bladders. The collected worms were placed in a solution of normal saline and fixed in hot AFA, then stored in 70% ethanol, stained with Semichon's acetocarmine and mounted in Canada balsam. Mounted specimens were measured and observed with light microscope and were illustrated with the aid of a camera lucida.

RESULTS

Gorgodera japonica Yamaguti, 1936
Synonyms:
Gorgodera japonica Yamaguti, 1936.
Gorgodera cygnoides: Seno, 1907.

Description of specimens collected in this

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* Corresponding author

study (Fig. 1) based upon 8 whole-mounted and fully matured worms. The dimensions of the worms are summarized in Table 1.

The body is elongate, tapering slightly posteriorly. The body-surface bears no spines and the body-wall is thin. The subterminal, round oral sucker is much smaller than the round ventral sucker, which is situated at about one sixth or seventh of the body-length from the anterior end. The sucker ratio is slightly above 1:2. There is no pharynx. The oesophagus is thick-walled and variable in length. The caeca reach into the posterior region of hind body and terminate beyond the posterior margin of the last testis (Fig. 1-A).

The nine testes are spherical to slightly elongate with irregular margins. The testes lie in two longitudinal rows, 5 in the right row and

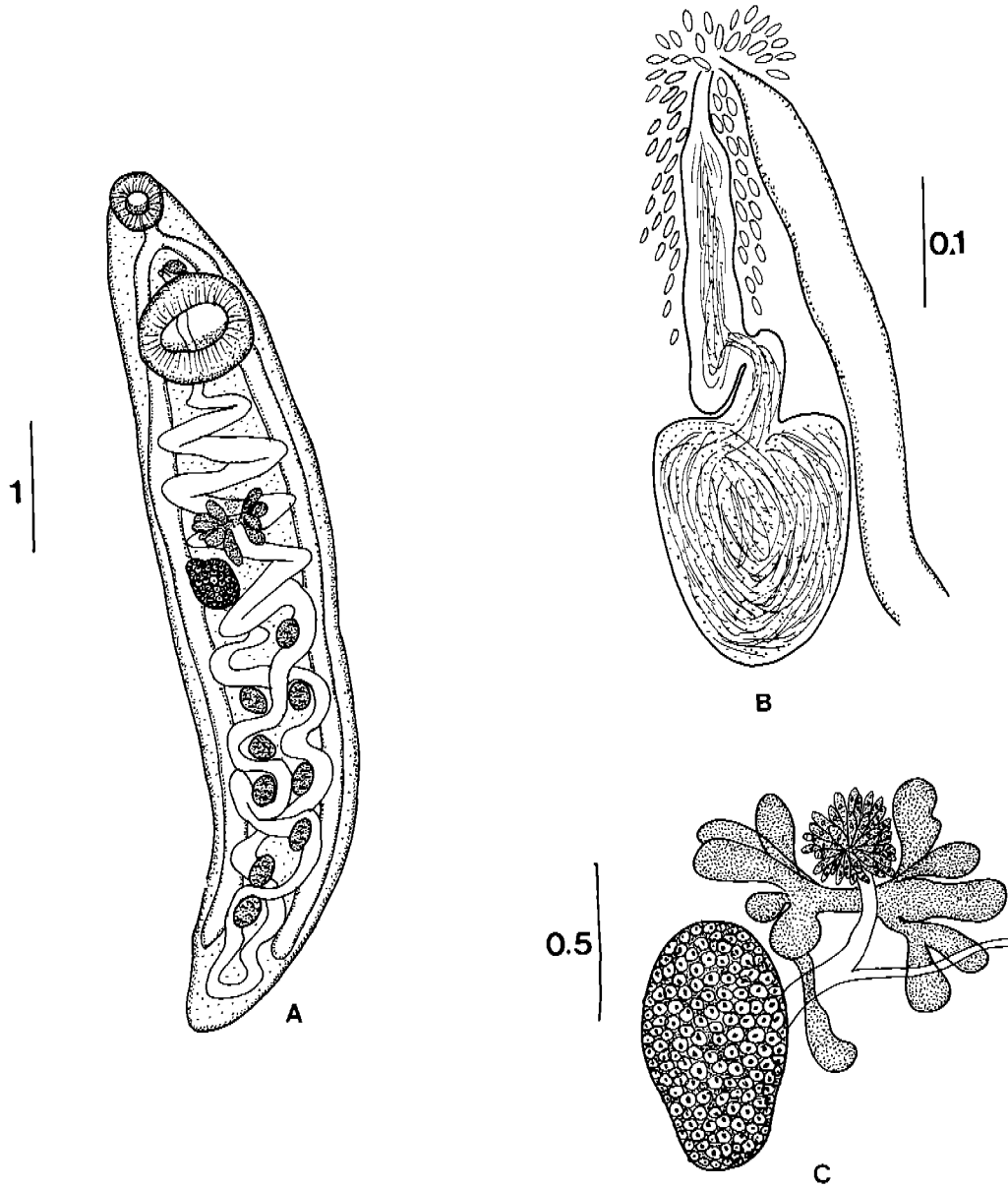

Fig. 1. *Gorgodera japonica* from *Rana nigromaculata* (A) fully mature specimen (B) seminal vesicle and metraterm (C) diagram of female proximal genitalia. Unit of bar scale = mm.

Table 1. Dimensions of *Gorgodera japonica* from the present material and from the original description

Authority	Yamaguti (1936)	Present material
Hosts	*Rana nigromaculata*	*Rana nigromaculata*
Locality	Japan	Korea
Length (mm)	3.5-5.5	4.0-9.3
Breadth (mm)	0.6-1.0	0.8-2.0
Oral sucker (mm)	0.2-0.325 in diameter	0.28-0.57 × 0.25-0.53
Ventral sucker (mm)	0.525-0.77 in diameter	0.65-1.13 × 0.59-1.13
Sucker-ratio	above two times	1:2.0-2.2
Oesophagus (mm)	0.2	0.10-0.25
Testes	nine in number two series	8-10 in number (usually 9) two series
Ovary (mm)	0.18-0.31 × 0.25-0.44	0.18-0.48 × 0.26-0.78
Vitellaria	paired rossette form each with 6-8 lobes	paired rosette form each with 6-9 lobes
Eggs (um)	40-44 × 27-31 (in life)	30-43 × 20-30 (in stained specimens)

4 in the left row, and are located in the intercaecal region of the hind body. They are variable in size usually smaller than the ovary and occasionally appear indistinct or absent. Some specimens have 8 (3/5) or 10 (5/5) testes. There is no cirrus pouch. The seminal vesicle is elongate to elliptical, located transversely or obliquely between the caecal bifurcation and the anterior margin of ventral sucker. The pars prostatica is tubular, slightly coiled, surrounded by gland cells that extend to the genital pore. The ejaculatory duct is short, narrow and cirrus is not seen (Fig. 1-B). The genital pore opens ventrally, just posterior to the caecal bifurcation.

The ovary varies in shape from elongate or pear shaped to elliptical, often with a large lobe. It lies directly posterior to the vitellaria, anterior to the first testis and slightly lateral to the median body line. The oviduct arises from the lateral surface of the ovary and receives the Laurer's canal and common vitelline duct. The Laurer's canal runs transversely to the opposite side of the ovary and opens dorsally. The Mehlis' gland lies between the two vitelline glands (Fig. 1-C). The vitellaria are paired, each pair forming a rosette with 6-9 lobes and lie posterior to the ventral sucker. The uterus is distributed mainly in the postacetabular region of the body, filling up all the available space of the hind body, with regular transverse loops between the posterior margin of the ventral sucker and the anterior margin of the ovary. The metraterm is tubular, slightly curved and narrowed at its distal portion. The numerous eggs are variable in size and shaped from spherical to elliptical. The egg shell is very thin and without operculum.

Gorgoderina bombinae Yu & Lee, 1983

Gorgoderina bombinae Yu & Lee, 1983

Description of specimens collected in this study (Fig. 2) based upon 5 whole-mounted specimens. The dimensions of the worms are summarized in Table 2.

The subcylindrical worms taper slightly at the posterior ends. The body surface is smooth. The subterminal, spherical oral sucker smaller than the round ventral sucker (ratio 1:1.3). The ventral sucker is situated at about one-quarter of the body length from the anterior extremity. There is no pharynx. The oesophagus is distinct and variable in length. The caeca do not reach the posterior extremity of body, but usually attain the posterior margin of the posterior testis (Fig. 2-A).

The testes are quite variable in shape, usually oval to elongate. The surface irregularity of testes is much severe in young adults. Occasionally, the testes of old adults are become very small or even absented. They are arranged obliquely tandem to directly tandem. The location of the testes is very variable. Usually in the older adults, they lie from anterior portion of the hind-body to middle of the hind-body, but in the younger adults, they lie only in posterior region of the hind-body (Fig. 2-B). There is no cirrus sac. The seminal vesicle is entirely oval to transversely elongate, often bipartite forms are

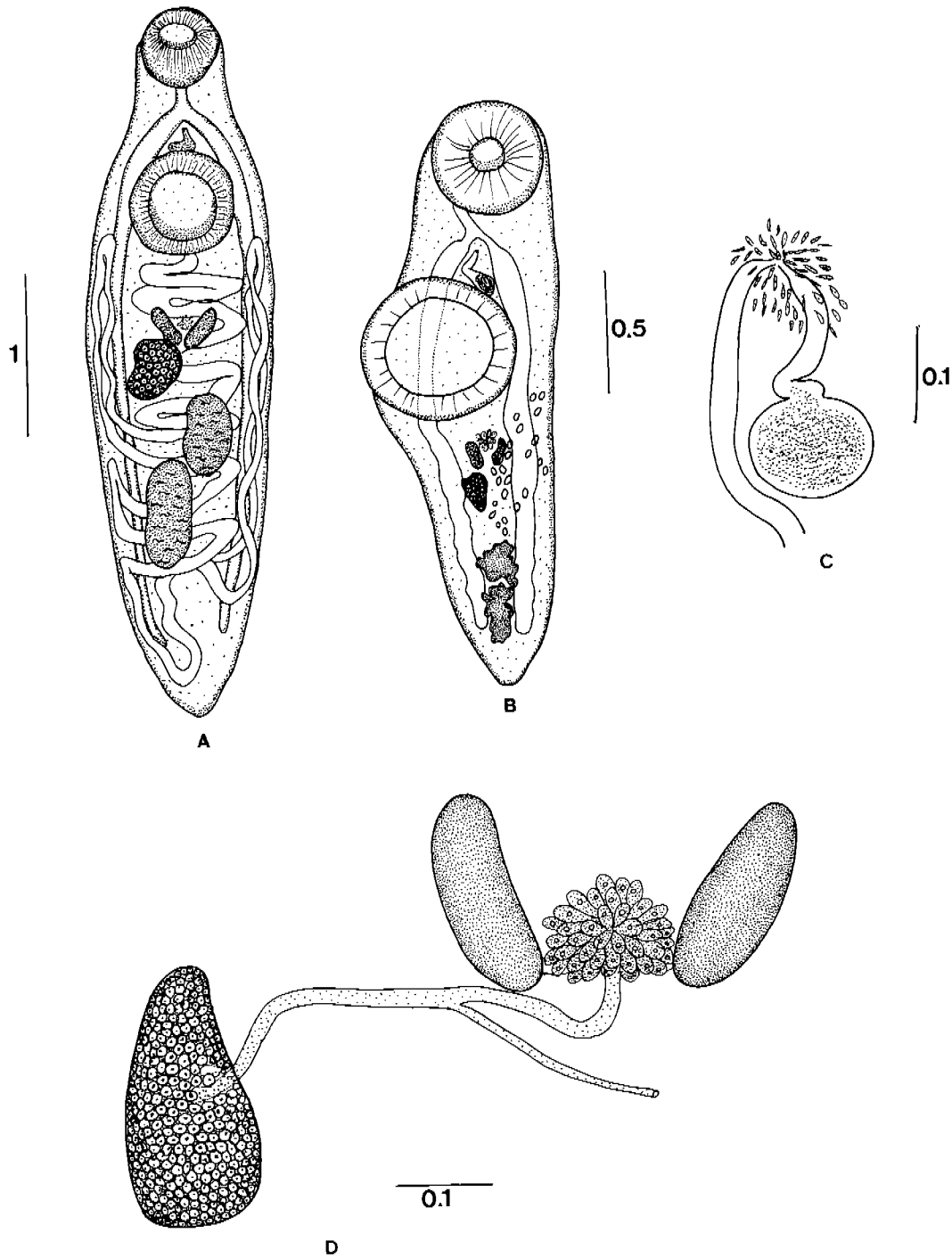

Fig. 2. *Gorgoderina bombinae* from *Bombina orientalis* (A) fully mature specimen (B) immature specimen (C) seminal vesicle and metraterm (D) diagram of female proximal genitalia. Unit of bar scale = mm.

found (Fig. 2-C). It lies just above the anterior margin of the ventral sucker. The pars prostatica is wide, somewhat coiled and surrounded by many gland cells. The

Table 2. Dimensions of *Gorgoderina bombinae* from the present material and from the original description

Authority	Yu & Lee (1983)	Present material
Host	*Bombina orientalis*	*Bombina orientalis*
Locality	Korea	Korea
Length (mm)	1.79-4.84	2.02-4.45
Breadth (mm)	0.41-1.13	0.50-1.07
Oral sucker (mm)	0.25-0.55 in diameter	0.40-0.48 × 0.43-0.47
Ventral sucker (mm)	0.38-0.66 in diameter	0.57-0.64 × 0.51-0.63
Sucker-ratio	1:1.2-1.5	1:1.26-1.40
Oesophagus (mm)	0.156-0.163	0.08-0.17
Testes (mm)	0.156-0.250 × 0.094-0.118	0.10-0.29 × 0.19-0.45
		0.10-0.25 × 0.17-0.63
Ovary (mm)	0.139-0.302 × 0.076-0.176	0.08-0.27 × 0.15-0.39
Vitellaria (mm)	3 in number	2 in number
	0.131-0.250	0.06-0.12 × 0.06-0.28
		0.05-0.14 × 0.09-0.30
Eggs (μm)	26-32 × 16-19	30-33 × 18-20

ejaculatory duct is short and narrower than the pars prostatica. The genital pore lies just behind the caecal bifurcation.

The ovary is irregular shape, usually reniform and slightly lobed. It is situated at just posterior to the vitellaria and a little out of the median body-line. The oviduct arises from inner side of the ovary and receives the Laurer's canal and the common vitelline duct. The Laurer's canal runs transversely. The oviduct is curved several times before receives the common vitelline duct (Fig. 2-D). The Mehlis' gland lies between or just in front of the vitellaria. The paired vitellaria are entire and elongated oval, lie relatively long distant from posterior margin of the ventral sucker. The uterus fills up all the space of the postacetabular region, intruding into the extracaecal fields. The metraterm is tubular and slightly curved. The numerous eggs are variable in size and shaped from globular to elliptical. The egg shell is very thin and without operculum.

DISCUSSION

Looss (1901) erected the family Gorgoderidae to include the subfamilies Gorgoderinae Looss, 1899 and Anaporrhutinae Looss, 1901. In the subfamily Gorgoderinae he included 2 genera, *Gorgodera* Looss, 1899 and *Phyllodistomum* Braun, 1899. In his earlier work Looss (1894) had mentioned that the frog bladder fluke described by Olsson as *Distomum vitellilobum* had only two testes instead of nine as in *Distomum cygnoides*, but he considered it to be a young form of the latter species. Later Looss (1902) divided the genus *Gorgodera*, and established for the species with two testes the genus *Gorgoderina*. The differences between the genus *Phyllodistomum* and *Gorgoderina* is not great and depends largely on differences in body shape, the members of the former genus being pyriform or spatulate, and the latter fusiform or lancet-like (Dollfus, 1958).

Prudhoe & Bray (1982) noted that *Gorgoderina* from amphibians and some *Phyllodistomum* from fishes and amphibians show a very close morphological resemblance to each other. Pande (1937) found non-spatulate body shaped gorgoderid worms in frogs and regarded *Gorgoderina* as a synonym of *Phyllodistomum*. Kaw (1950) and Frandsen (1957) considered them to be synonymous, also. Although the hind-body of living specimens of *Phyllodistomum* may sometimes assume the lanceolate appearance commonly found in *Gorgoderina*, when fixed the hind-body of *Phyllodistomum* becomes somewhat discoid. Furthermore, in *Gorgoderina* the ventral sucker has a tendency to lie nearer to the oral sucker than to the middle of the body, to be distinctly larger than the oral sucker, and to have a diameter frequently more than half

the maximum width of the body, even sometimes greater than the width (Prudhoe & Bray, 1982).

Ozaki (1926) erected the genus *Microlecithus* and reported *Microlecithus kajika* n.sp., which found in the urinary bladder of *Polypedates buergeri* (Schelegel) from Japan. Later Ozaki (1935) transferred it to the genus *Gorgoderina* . Yamaguti (1934) and Bhalerao (1937) classified *Microlecithus* as a synonym of *Phyllodistomum*. The genus *Microlecithus* is, however, closer to *Gorgoderina* than *Phyllodistomum* in that ventral sucker lie nearer to the oral sucker, distinctly larger than the oral sucker and have a diameter more than half the maximum width of the body (Prudhoe and Bray, 1982). According to the original description and figure, the body shape of *M. kajika* is lanceolate form. Therefore it is proper that *Microlecithus* should be classified as a synonym of *Gorgoderina*.

As far as the systematics of the subfamilies in Gorgoderidae are concerned, there have been many problems in weighing the relative values of different characters. The number of testes has been used as a fundamental value in distinguishing subfamilies (Pigulevsky, 1953), but on the other hand the shape of body has also been considered as a key character in dividing subfamilies (Dawes, 1946; Yamaguti, 1958).

Fotedar (1969) refuted Yamaguti's (1958) classification as an unnatural division and placed *Gorgoderina* and *Phyllodistomum* under the subfamily Phyllodistomatinae Pigulevsky, 1953. Indeed the studies on the excretory system suggest a very close relationship between *Phyllodistomum* and *Gorgoderina* (Byrd *et al.*, 1940).

On the contrary, Rankin (1939) emphasized that *Gorgodera* and *Gorgoderina* are very closely related morphologically and also in respect to their life cycles and modes of development. According to Rankin's results metacercariae of *Gorgoderina attenuata* have nine testes and immature individuals show the gradual fusion to two, as found in adult worm. If these facts are confirmed, this may indicate that *Gorgoderina* has arisen from *Gorgodera*, not *Phyllodistomum* (Brooks & MacDonald, 1986). Mitchell (1973), however, reported that the testes of the earliest juvenile *Gorgoderina vitelliloba* are usually just discernible as small clusters of cells and Rankin's results, therefore, should be reexamined.

From above respects, it would be premature and artificial to classify subfamilies, so we did not applicate subfamily level in classification of Gorgoderidae.

Pigulevsky (1953) classified the genus *Gorgodera* into five subgenera- *Antodera, Extremodera, Mediodera, Postodera, Gorgodera*. Because the characters used to distinguish these subgenera were somewhat artificial and variable even in the same species according to the developmental stages, Prudhoe and Bray (1982) did not accept Pigulevsky's classification and we also quite agree with their opinion.

Pereira and Cuocolo (1940) divided *Gorgoderina* into two subgenera (*Gorgoderina, Neogorgoderina*) according to the arrangement of the vitelline glands. There is, however, contradiction in type-species, therefore the subgenera proposed by Pereira and Cuocolo are not accepted. Pigulevsky (1952) classified *Gorgoderina* into two subgenera, *Gorgoderina* and *Gorgoderimma*, on the basis of the distance between ventral sucker and vitelline glands. Yamaguti (1958) classified *Gorgoderina* without subgeneric division and this classification scheme is accepted here because of the uncertainty and confusion associated with the subgeneric division.

Yamaguti (1936) reported *Gorgodera japonica* as a new species and described differences from closely related species *G. cygnoides*, chiefly in the size of the body and eggs. He suggested that *G. cygnoides* reported by Seno (1907) was probablely *G. japonica*. Pigulevsky (1952) distinguished between *G. cygnoides* and *G. japonica* according to their sucker ratios, i. e., the ventral sucker of *G. japonica* is two times larger than oral sucker and the ventral sucker of *G. cygnoides* is less than two times larger than oral sucker. The sucker ratio of our specimens fall under *G. japonica* according to Pigulevsky, however, several authors (Harwood, 1932; Goodchild,1948) pointed out that sucker ratio ranges were variable and of little specific significance in the gorgoderid group except in the fully matured individuals. We believe that the character distinguishing *G. japonica* from

G. cygnoides is not the size of the body and the sucker ratio but deeply branched vitellaria in *G. japonica*. Based on that character, we agree with Yamaguti (1936) that *G. cygnoides* reported by Seno (1907) must be corrected as *G. japonica*.

In the original description of *Gorgoderina bombinae*, Yu & Lee (1983) described the number of vitellaria as three, but through our observation of those type and paratype specimens loaned by professor Lee that description must be corrected to two.

Two species of *Gorgoderina*, *G. tanigawaensis* Uchida & Itagaki, 1974 and *G. kajika* (Ozaki, 1926), are recorded from Japan.

Gorgoderina bombinae can be distinguished from *G. tanigawaensis* in the smaller body size and in having non-lobed testes and vitellaria. *G. kajika* is very similar to the present species in many characters, but has smaller eggs and larger testes than *G. bombinae*.

G. bombinae differs from *G. vitelliloba* (Olsson, 1876) in having compact, entire vitellaria and smaller eggs; from *G. skrjabini* Pigulevsky, 1953 in having entire vitellaria and larger eggs.

G. parvicava Travassos, 1922 is distinguished by the fact that its oral sucker is larger than ventral sucker. *Gorgoderina bombinae* differs from *G. tanneri* Olsen, 1937; *G. attenuata* (Stafford, 1902); *G. capsensis* Joyeux & Baer, 1934; *G. aurora* Ingles, 1936 in having sucker ratios less than 1:2. It differs from *G. multilobata* Ingles & Langston, 1933; *G. translucida* (Stafford, 1902), *G. tenua* Rankin, 1937; *G. bilobata* Rankin, 1937; *G. schistorchis* Steelman, 1938; *G. guptai* Jahan, 1973; *G. permagna* Lutz, 1926 in having non-lobed vitellaria.

Finally, *G. bombinae* differs from *G. ellipticum* Dwivedi, 1968; *G. diaster* Lutz, 1926; *G. cedroi* Travassos, 1924; *G. cryptorchis* Travassos, 1924; *G. carli* Baer, 1930; *G. symmetriorchis* Dwivedi, 1968; *G. infundibulata* Dwivedi, 1968 in having vitellaria are not located directly posterior to the ventral sucker.

In conclusion, *G. bombinae* is distinguished from the other species by the following characters; the sucker ratio, the vitellaria shape and location, the egg size and body shape.

While the number of testes may have significance at the generic level, this character seems to be variable. In some worms of *G. japonica* and *G. bombinae* of our specimens, the testes are lacked or vestigial. In *Gorgoderina attenuata*, it was noted that many worms lacked testes and in a few no vitelline glands could be found (Nollen and Alberico, 1972), also. Nollen and Alberico also attempted to correlate the loss of the testes with various factors and found that aging and poor nutrition of the host were strongly correlated with degenerative changes in testes numbers. According to Nollen and Alberico's results, the degenerative variations of the testes in these two species can be explained. The more testes (5/5) in *G. japonica*, however, can not be explained by those results. The genetic approaches are needed to elucidate this phenomenon.

Rana nigromaculata was the only recorded host of *G. japonica* (Yamaguti, 1936; Song *et al.*, 1965; Lee *et al.*, 1976; Yu & Lee, 1983) and is our specimens also. And *G. bombinae* is recovered only from *Bombina orientalis*. The host specificities of other gorgoderid species are not strictly restricted to the one host species. The host specificity of *G. japonica* and *G. bombinae*, however, is most high in Korea.

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=초록=

한국산 개구리류의 방광에 기생하는 Gorgoderid 흡충류의 분류

김기홍[1], 주경환[2], 임한종[2]

부산수산대학교 수산과학대학 어병학과[1], 고려대학교 의과대학 기생충학교실 및 열대풍토병연구소[2]

Gorgoderidae에 속하는 흡충류는 어류, 양서류 및 파충류 등의 방광에 기생하며, 우리 나라의 양서류에서는 *Gorgodera japonica* 1종만이 기록되어 있다가 유와 이(1983)에 의해 *Gorgoderina bombinae* 1종이 신종으로 보고된 바 있다. 본 연구에서는 1989년부터 1994년에 걸쳐 전국 각지에서 채집한 양서류를 대상으로 그들의 방광에 기생하는 윤충류를 조사한 결과 *Gorgodera japonica* 및 *Gorgoderina bombinae* 2종을 동정하였으며, 분류된 2종에 대한 형태학적인 특징을 기술하고 이들의 분류학적 위치에 대해 상세히 논하였다. *G. japonica*가 *G. cygnoides*와 명확히 구별되는 점은 난황선이 깊은 열개에 의해 뚜렷한 여러 개의 엽을 형성하는 점이었다. *G. bombinae*는 기존의 종들과 비교한 결과 뚜렷한 종으로서 확인되었으며, 이 종에 대한 국명을 "무당개구리방광흡충"으로 제안한다.

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