

## Amphistomes of Vietnamese vertebrates (Trematoda: Amphistomida)

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ABSTRACT. An examination of the amphistomes of Vietnamese vertebrates, based on original test material, revealed 28 species. Four of them were found in fishes, two in amphibians and 22 in mammals. Surface topography, histomorphological structure of the muscular organs of certain species have been studied and a parasite/host and a host/parasite check-list have been compiled.

KEY WORDS. Amphistomes, Vietnamese vertebrates, scanning electron microscopy, histomorphology of muscular organs, parasite/host and host/parasite check-lists.

Reviewing relevant literature data referring to the amphistomes of Viet Nam or the neighbouring area previously called Indochina, one experiences that varying interest has been rayed to the study of amphistomes of lower and higher vertebrates. While numerous reports have been published on the amphistomes of mammals (BARROIS, 1908; RAILLIET, 1924; HOUEMER, 1938; DOLLFUS, 1963; DROZDZ and MALCZEWSKI, 1967; SEGAL et al., 1968; THAN THÉ VIET et al., 1977; SEY, 1980, 1983, 1985), there are few on the amphistomes of fishes and amphibians (HA KI, 1969; ODENING, 1968; SEY, 1986a). These studies revealed 42 species: Amurotrema dombrowkajae Achmerow, 1959; Neocladorchis multilobularis Sey, 1986; Platycladorchis microacetabularis Sey, 1986; P. macroacetabularis Sey, 1986; Diplodiscus amphichrus Tubangui, 1938; D. mehrai Pande, 1937; Watsonius noci (Barrois, 1908); Hawkesius hawkesi (Cobbold, 1875); Homalogaster paloniae Poirrier, 1883; Gastrodiscoides hominis (Lewis et McConnel, 1876); Paramphistomum cervi (Zeder, 1790); P. epiclitum Fiscoeder, 1904; P. ichikawai Fukui, 1922; P. gotoi Fukui, 1922; P. liorchis Fiscoeder, 1901; Calicophoron calicophorum (Fiscoeder, 1901); C. microbothrioides (Price et McIntosh, 1944); C. cauliorchis (Stiles et Goldberger, 1910); C. ijimai (Fukui, 1922); C. papillosum (Stiles et Goldberger, 1910); Orthocoelium scoliocoelium (Fiscoeder, 1901); O. orthocoelium (Fiscoeder, 1901); O. dicranocoelium (Fiscoeder, 1901); O. streptocoelium (Fiscoeder, 1901); O. dinniki Eduardo, 1982; O. saccocoelium Sey, 1980; Cotylophoron cotylophorum (Fiscoeder, 1901); C. indicum Stiles et Goldberger, 1910; Explanatum explanatum (Creplin, 1847); E. anisocotyle (Faust, 1920); E. bathycotyle (Fiscoeder, 1901); Gigantocotyle formosana (Fukui, 1929); G. fraternum (Stiles et Goldberger, 1910); Carmyerius spatiosus (Brandes, 1898); C. bulbosus Sey, 1985; C. synethes (Fiscoeder, 1901); Gastrothylax crumenifer (Creplin, 1847); G. glandiformis Yamaguti, 1939; G. minutus Fiscoeder, 1901; Fiscoederius cobboldi (Poirrier, 1883); F. elongatus (Poirrier, 1833); F. japonicus Fukui, 1922. With a few exceptions (SEY, 1983, 1985) these examinations were mainly based on gross morphological observations, whereas it is generally accepted that modern diagnosis of amphistomes cannot be accomplished without a knowledge of the structure of the muscular organs.

Our examinations, which have also been extended to histomorphological traits, confirmed, on the one hand, the occurrence of the majority of the previously described species, and on the

other hand further new information on the amphistomes of Vietnamese vertebrates are presented.

#### MATERIALS AND METHODS

The majority of the test material of fishes, amphibians and turtles (Testudo elongata, T. impressa, Trionyx steindachneri) were collected from hosts deposited in formaline in the Department of Vertebrate Zoology, Hanoi University, and the lesser part of it was bought at the Hanoi market. The test material of mammalian hosts is deposited at the Department of Parasitology, Vietnamese Scientific Research Center, Hanoi, collected from Bos primigenius f. taurus, Bubalus arnee f. bubalis, Capra aegargus f. hircus, Cervus unicolor, Muntiacus muntjak and Sus crofa f. domestica. Whole mounts were stained in carmine, median sagittal sections were prepared by the usual method and the technique applied for examination of surface topography was the same as described by the author elsewhere (SEY, 1985b).

#### RESULTS AND DISCUSSION

Amphistomes were recovered from all of the vertebrate groups except reptiles (turtles).

##### AMPHISTOMES OF FISHES

Helminth parasites of freshwater fishes have been infrequently investigated (OSHMARIN, 1965; HA KI, 1969; SEY, 1969a). Of the trematodes described, one species of amphistome, Amurotrema dombrowskajae was recorded by HA KI (1969) from Spinibarichthys denticulatus. I have found this species in the same host. Histomorphological examination of its muscular organs showed that it had a new type of pharynx, Amurotrema (characterized by primary pharyngeal sacs longer than the pharynx itself, as well as by the presence of a moderately developed middle circular layer, Fig. 13). The acetabulum is of a Cleptodiscus-type (SEY, 1986b).

Further three fish amphistomes (Neocladorchis multilobularis, Platycladorchis microacetabularis, P. macroacetabularis) have been described elsewhere (SEY, 1986a).

##### AMPHISTOMES OF AMPHIBIANS

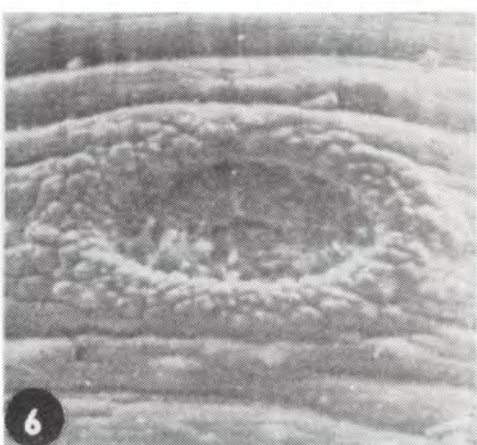
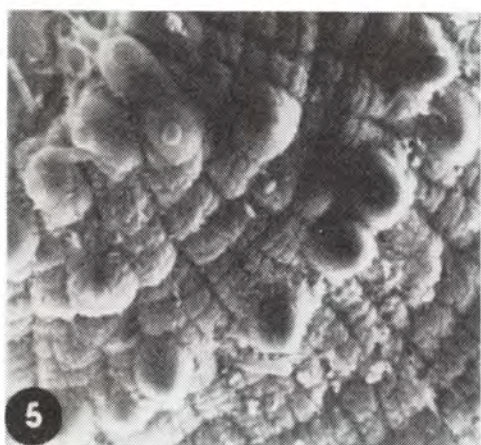
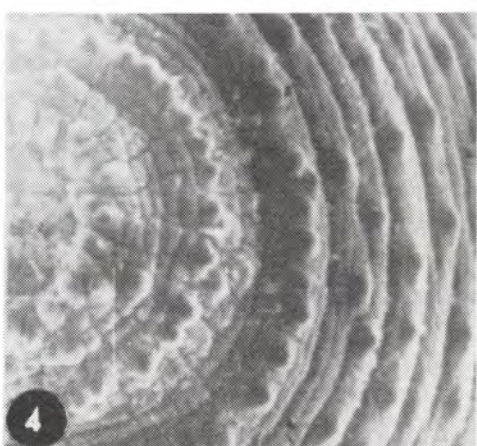
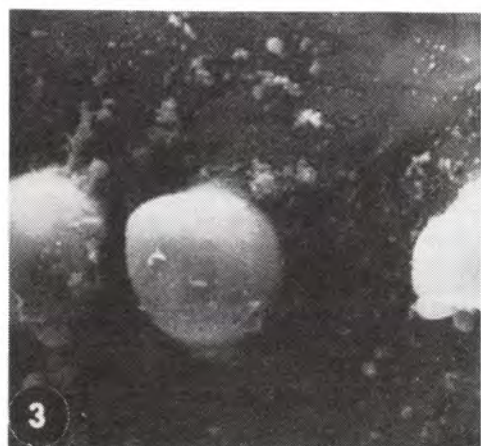
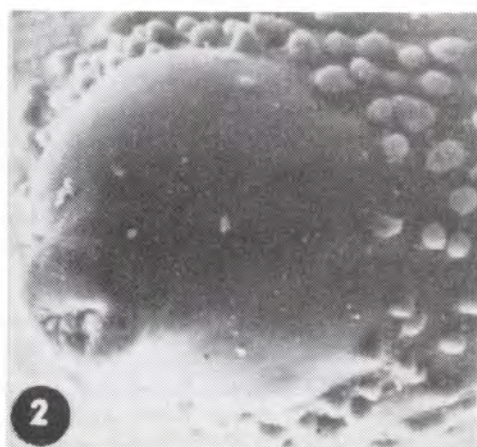
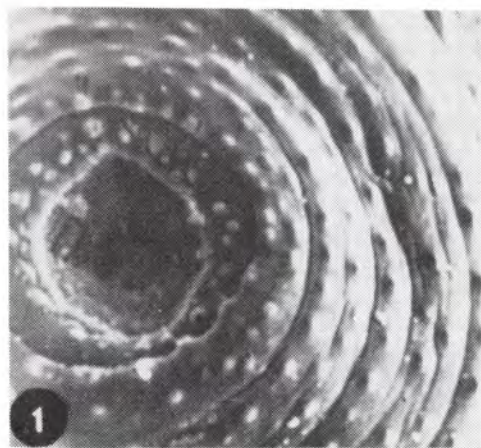
Two species of amphistomes (Diplodiscus amphichrus Tubangui, 1933 and D. mehrai Pande, 1937) were found in frogs and salamanders. The former species was described for the first

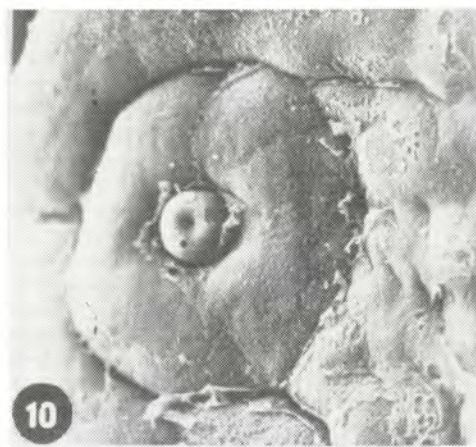
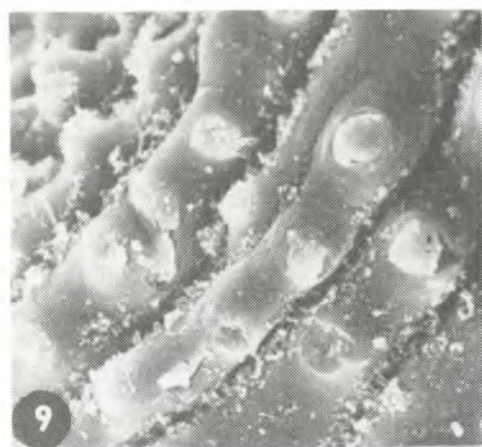
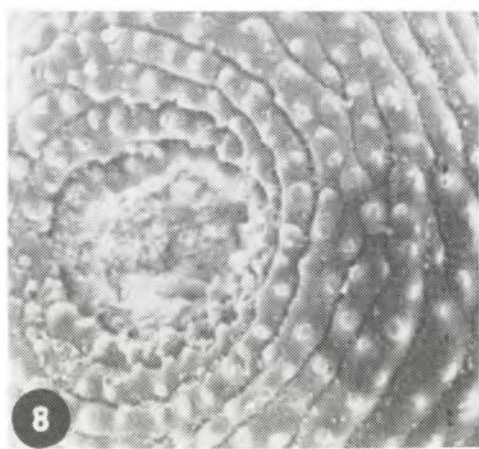
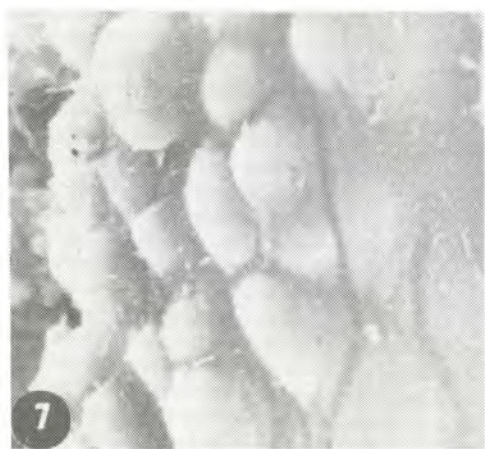
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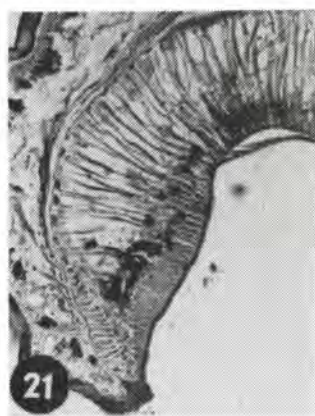
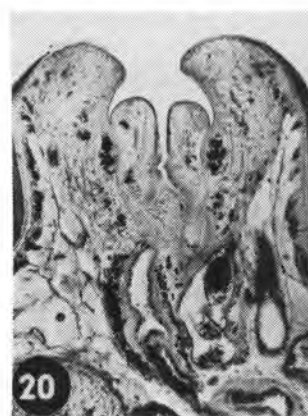
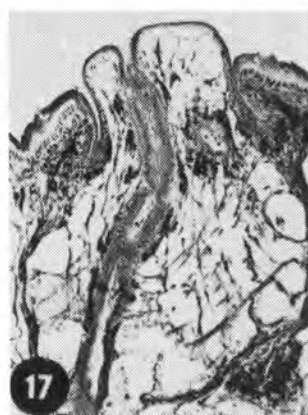
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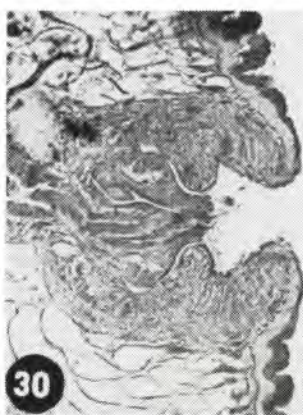
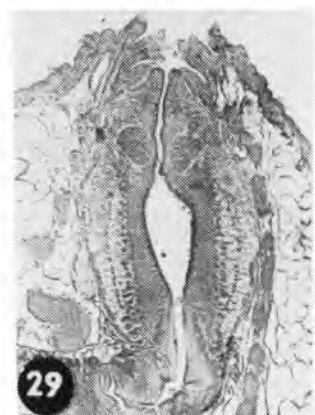
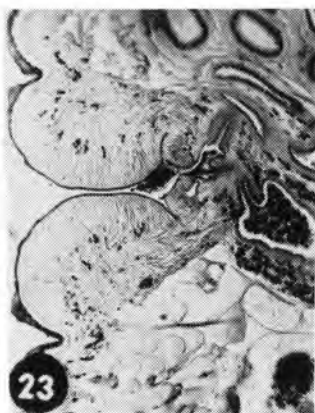
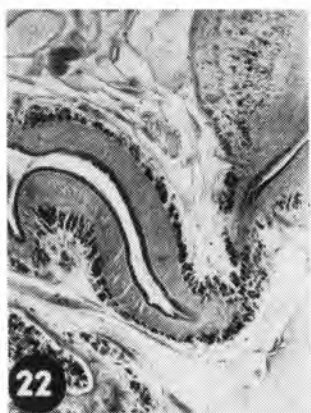
Figs 1-12: Scanning electron microscopy of surface topography: 1-3 Gastrodiscoides hominis (1= general view of oral opening, X54; 6-7 genital opening, 6= X190, 7= X695); 4-7 Orthocoelium saccocoelium (4-5 general view of oral opening, 4= X81, 5= X196, 6-7 genital opening, 6= X85, 7= X180); 8-10 Gastrothylax crumenifer (8-9 general view of oral opening, 8= X78, 9= X266, 10= genital opening, X73); 11-12 Carmyerius synethes (general view of oral opening, 11= X186, 12= X990).

Figs 13-30: Median sagittal sections of muscular organs: Amurotrema dombrowskajae (13= pharynx), Diplodiscus amphichrus (14= pharynx), D. mehrai (15= pharynx), Hawkesius hawkesi (16= pharynx, 17= genital opening, 18= dorsal and 19= ventral halves of acetabulum), Paramphistomum ichikawai (20= genital opening, 21= dorsal half of acetabulum), Calicophoron microbothrioides (22= oesophagus, 23= genital opening, 24= dorsal half of acetabulum), Orthocoelium dinniki (25= oesophagus, 26= genital opening), O. saccocoelium (29= pharynx, 30= genital opening, 27= dorsal and 28= ventral half of acetabulum).









time by ODENING (1968) from Rana tigrina rugulosa and the latter one was recovered from Rana limnocharis and Paramesotriton deloustali. The findings of D. mehrai represent new hosts and locality records.

Since a great number of specimens were available for study, it was possible to examine the structure of the muscular organs in both species. These examinations revealed a solid ground for the validity of the species in question and lead to a discovery of the specific traits which can be used in their differentiation. Accordingly, the structure of the pharynx (Subclavatus in D. amphichrus, Fig. 14; and Megalodiscus in D. mehrai, Fig. 15), the position of the genital opening (post-bifurcal in D. amphichrus and bifurcal in D. mehrai) and the arrangement of the vitelline follicles (discontinuous at the middle part of each lateral region in D. amphichrus and continuous in D. mehrai) are the most important specific characters.

#### AMPHISTOMES OF MAMMALS

Until recently 36 species of amphistomes have been shown to occur in mammals. We have, however, found the following 22 species in the samples available for examination: Homalogaster paloniae, Hawkesius hawkesi, Gastrodiscoides hominis, Paramphistomum epiclitum, P. gotoi, P. ichikawai, Calicophoron calicophorum, C. papillosum, C. microbothrioides, Explanatum explanatum, Gigantocotyle formosanum, Orthocoelium dicranocoelium, O. dinniki, O. saccocoelium, O. scolicoelium, Gastrothylax crumenifer, Carmyerius bulbosus, C. spatiosus, C. synthes, Fischoederius cobboldi, F. elongatus and F. japonicus.

Of the species discovered on the basis of our examinations Paramphistomum ichikawai, Calicophoron microbothrioides, Orthocoelium saccocoelium and O. dinniki were described for the first time from Viet Nam, thus represent new locality records.

The pouched amphistomes found in Vietnamese ruminants have been described elsewhere (SEY, 1985a).

An examination of the surface topography of the species, Gastrodiscoides hominis (Figs 1-3), O. saccocoelium (Figs 4-7), Gastrothylax crumenifer (Figs 8-10) and Carmyerius synthes (Figs 11-12) by scanning microscopy revealed three types of papillae (dome to conical non-ciliated papilla, Figs 7, 9), crateriform papilla (Figs 11, 12); short and stumpy papilla, sitting on a tegumental elevation (Fig. 5), which are similar to that described previously by EDUARDO (1982) and SEY (1984b).

Of the species recorded for the first time in Viet Nam, Paramphistomum ichikawai has a Calicophoron-type of pharynx, an Ichikawai-type of genital opening (Fig. 20) and a Pisum-type of acetabulum (Fig. 21); the same organs in Calicophoron microbothrioides are: Calicophoron with oesophagus having muscular thickening (Fig. 22), Microbothrium (Fig. 23) and Pisum (Fig. 24); in Orthocoelium saccocoelium: Saccocoelium (Fig. 29), Papillogenitalis (Fig. 30) and Streptocoelium (Figs 27-28), in O. dinniki: Calicophoron with oesophagus having muscular thickening along the posterior half (Fig. 25), Papillogenitalis (Fig. 26) and Streptocoelium.

The samples of Hawkesius hawkesi rendered an examination of the structure of the muscular organs possible for the first time for a this species. The pharynx is characterized by the presence of a moderately developed pharyngeal bulb and well-developed secondary pharyngeal sacs. The pharynx itself is poorly developed and similar to that of the Homalogater (SEY, 1984) and Pseudodiscus (NASMARK, 1937) types, but differs from them by having a moderately developed middle circular and well-developed inner longitudinal layers. I regard this as a new type, named Hawkesius (Fig. 16).

The genital opening is moderately developed, characterized by the presence of genital papillae, absence of circular musculature and the existence of a definitive fibre layer, delimitating the genital opening from the surrounding tissue, like to genital sucker. There is a short hermaphrodite duct. It represents a new type of genital opening, named Hawkesius (Fig. 17).

It is most similar to the Balanorchis-type (EDUARDO, 1982) but differs from it in the absence of sphincter papillae.

The acetabulum is characterized by a poorly developed marginal longitudinal muscle layer and the number of muscle units found in DE (42-45, Fig. 18) and VE (22-25, Fig. 19). It is similar to that of the Gastrodiscoides (SEY, 1986b) and the Homalogaster-types but differs from them in having lesser muscle units in the DE and VE muscle layers. It is regarded as a new type, named Hawkesius.

Of the amphistomes previously described from Viet Nam, fourteen species (Watsonius noci, Paramphistomum cervi, P. liorchis, Calicophoron cauliorchis, C. ijimai, Orthocoelium orthocoelium, O. streptocoelium, Cotylophoron cotylophorum, C. indicum, Gigantocotyle anisocotyle, G. bathycotyle, G. fraternum, Gastrothylax glandiformis and G. minutus) have not been revealed during our examinations. The clarification of their real position, presence or absence should be the subject of further studies.

#### A KEY TO SPECIES OF VIETNAMESE AMPHISTOMES

- |  |  |
|--|--|
| 1) Ventral pouch absent .....  | 2  |
| - Ventral pouch present .....  | 14                                       |
| 2) Pharynx with primary pharyngeal sacs .....  | 3  |
| - Pharynx with pharyngeal bulb and secondary pharyngeal sacs .....                             | 7  |
| - Pharynx without primary pharyngeal sacs, pharyngeal bulb and secondary pharyngeal sacs ..... | 8  |
| 3) Parasites of fishes .....   | 4  |
| - Parasites of amphibians .....  | 6  |
| 4) Cirrus pouch normally developed .....   | 5  |
| - Cirrus pouch strongly developed .....  | <i>Platycladorchis microacetabularis</i> |
| 5) A) Testes spherical .....   | <i>Amurotrema dombrowskajae</i>          |
| B) Testes strongly lobed .....   | <i>Neocladorchis multilobularis</i>      |
| C) Testes with some lobes, acetabulum big .....  | <i>Platycladorchis macroacetabularis</i> |
| 6) A) Pharynx Subclavatus-type .....   | <i>Diplodiscus amphichrus</i>            |
| B) Pharynx Megalodiscus-type .....   | <i>Diplodiscus mehrai</i>                |
| 7) A) Genital opening Noci-type .....  | <i>Watsonius noci</i>                    |
| B) Genital opening Homalogaster-type .....   | <i>Homalogaster paloniae</i>             |
| C) Genital opening Parvipapillatum-type .....  | <i>Gastrodiscoides hominis</i>           |
| D) Genital opening Hawkesius-type .....  | <i>Hawkesius hawkesi</i>                 |
| 8) Pharynx Calicophoron-type .....   | 9  |
| - Pharynx Dicranocoelium-type .....  | 12                                       |
| - Pharynx Explanatum-type .....  | 13                                       |
| - Pharynx Liorchis-type .....  | <i>Paramphistomum gotoi</i>              |
| 9) Genital opening Papillogenitalis .....  | 10                                       |
| - Genital opening other type .....   | 11                                       |
| 10) A) Acetabulum Streptocoelium-type .....  | <i>Orthocoelium dinniki</i>              |
| B) Acetabulum Calicophoron-type .....  | <i>Calicophoron papillosum</i>           |
| 11) A) Genital opening Ichikawai-type .....  | <i>Paramphistomum ichikawai</i>          |
| B) Genital opening Epiclitum-type .....  | <i>Paramphistomum epiclitum</i>          |
| C) Genital opening Calicophoron-type .....   | <i>Calicophoron calicophorum</i>         |
| D) Genital opening Microbothrium-type .....  | <i>Calicophoron microbothrioides</i>     |
| 12) A) Genital opening Scoliocoelium-type .....  | <i>Orthocoelium scoliocoelium</i>        |
| B) Genital opening Gracile-type .....  | <i>Orthocoelium dicranocoelium</i>       |



- 13) A) Genital opening Explanatum-type ..... Explanatum explanatum  
 B) Genital opening Microbothrium-type ..... Gigantocotyle formosanum
- 14) Uterus in dorsal median field along its length ..... 15  
 - Uterus crossing from one side of body to other near middle ..,Gastrothylax crumenifer
- 15) Testes symmetrical, one on each side of median line ..... 16  
 - Testes tandem in median line ..... 18
- 16) A) Oesophagus without oesophageal musculature ..... 17  
 B) Oesophagus with oesophageal thickening .....Carmyerius bulbosus
- 17) A) Genital opening Gracile-type ..... Carmyerius spatiosus  
 B) Genital opening Synthes-type ..... Carmyerius synthes
- 18) A) Genital opening Elongatus-type .....Fischoederius elongatus  
 B) Genital opening Microbothrium-type .....Fischoederius cobboldi  
 C) Genital opening Microbothriopapillatus-type .....Fischoederius japonicus

PARASITE/HOST CHECK-LIST

Amurotrema dombrowskajae  
 Spinibarbichthys denticulatus

Neocladorchis multilobularis  
 Spinibarbichthys denticulatus

Platycladorchis microacetabularis  
 Spinibarbichthys denticulatus

Platycladorchis macroacetabularis  
 Lissochilus krempfi

Diplodiscus amphichrus  
 Rana tigrina rugulosa

Diplodiscus mehrai  
 Rana limnocharis  
 Paramesotriton deloustali

\*Watsonius noci  
 Macaca mulatta

Homalogaster paloniae  
 Bos primigenius f. taurus  
 Capra aegargus f. hircus  
 Muntiacus muntjak

Gastrodiscoides hominis  
 Sus crofa f. domestica  
 Homo sapiens

Hawkesius hawkesi  
 Elephas maximus

Paramphistomum gotoi  
 Bubalus arnee f. bubalis

Paramphistomum ichikawai  
 Bubalus arnee f. bubalis

\*Paramphistomum cervi  
 Bos primigenius f. taurus  
 Bubalus arnee f. bubalis

Paramphistomum epiclitum  
 Bos primigenius f. taurus  
 Bubalus arnee f. bubalis

\*Paramphistomum liorchis  
 Bos primigenius f. taurus  
 Bubalus arnee f. bubalis  
 Capra aegargus f. hircus

Calicophoron calicophorum  
 Bos primigenius f. taurus  
 Bubalus arnee f. bubalis  
 Capra aegargus f. hircus  
 Cervus unicolor  
 Muntiacus muntjak

Calicophoron papillosum  
 Bubalus arnee f. bubalis  
 Bos primigenius f. taurus

\*Calicophoron cauliorchis  
 Capra aegargus f. hircus

\*Calicophoron ijimai  
 Bos primigenius f. taurus

Calicophoron microbothrioides  
 Bos primigenius f. taurus  
 Cervus unicolor  
 Muntiacus muntjak

Orthocoelium scolioceelum  
 Bos primigenius f. taurus  
 Bubalus arnee f. bubalis  
 Capra aegargus f. hircus  
 Muntiacus muntjak

\*Orthocoelium orthocoelium  
 Bos primigenius f. taurus  
 Bubalus arnee f. bubalis

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 \*Refers to literature data.

Orthocoelium dicranocoelium  
Bos primigenius f. taurus

Orthocoelium dinniki  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis  
Cervus unicolor

Orthocoelium saccocoelium  
Cervus unicolor

\*Orthocoelium streptocoelium  
Bubalus arnee f. bubalis

\*Cotylophoron cotylophorum  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis

\*Cotylophoron indicum  
Bos primigenius f. taurus

Explanatum explanatum  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis

Gigantocotyle formosanum  
Bubalus arnee f. bubalis

\*Gigantocotyle anisocotyle  
Bubalus arnee f. bubalis

\*Gigantocotyle bathycotyle  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis

\*Gigantocotyle fraternum  
Bubalus arnee f. bubalis

Gastrothylax crumenifer  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis  
Capra aegargus f. hircus  
Ovis ammon f. aries

\*Gastrothylax glandiformis  
Bubalus arnee f. bubalis  
Capra aegargus f. hircus

\*Gastrothylax minutus  
Bubalus arnee f. bubalis

Carmyerius spatiosus  
Bubalus arnee f. bubalis  
Bos primigenius f. taurus  
Muntiacus muntjak

Carmyerius bulbosus  
Bubalus arnee f. bubalis

Carmyerius synthes  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis  
Capra aegargus f. hircus  
Cervus unicolor

Fischoederius cobboldi  
Bos primigenius f. taurus

Fischoederius elongatus  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis  
Muntiacus muntjak

Fischoederius japonicus  
Bos primigenius f. taurus  
Bubalus arnee f. bubalis  
Cervus unicolor  
Muntiacus muntjak

#### HOST/PARASITE CHECK-LIST

Lissochilus krempfi  
Platycladorchis macroacetabularis

Spinibarbichthys denticulatus  
Platycladorchis microacetabularis  
Amurotrema dombrowskajae  
Neocladorchis multilobularis

Rana tigrina rugulosa  
Diplodiscus amphichrus

Rana limnocharis  
Diplodiscus mehrai

Paramesotriton deloustali  
Diplodiscus mehrai

\*Macaca mulatta  
Watsonius noci

\*Homo sapiens  
Gastrodiscoides hominis

Sus crofa f. domestica  
Gastrodiscoides hominis

Elephas maximus  
Hawkesius hawkesi

Bos primigenius f. taurus  
Homalogaster paloniae  
Paramphistomum cervi  
P. epiclitum

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\*Refers to literature data.

Bos primigenius f. taurus (cont.)

P. liorchis  
Calicophoron calicophorum  
C. papillosum  
C. ijimai  
C. microbothrioides  
Explanatum explanatum  
Orthocoelium scolioceolium  
O. orthocoelium  
O. dicranocoelium  
O. dinniki  
Cotylophoron cotylophorum  
C. indicum  
Gigantocotyle bathycotyle  
G. fraternum  
Gastrothylax crumenifer  
Carmyerius spatiosus  
C. synethes  
Fischoederius cobboldi  
F. elongatus  
F. japonicus

Bubalus arnee f. bubalis

Paramphistomum gotoi  
P. ichikawai  
P. cervi  
P. epiclitum  
P. liorchis  
Calicophoron calicophorum  
C. papillosum  
Orthocoelium scolioceolium  
O. orthocoelium  
O. dinniki  
O. streptocoelium  
Cotylophoron cotylophorum  
Explanatum explanatum  
Gigantocotyle formosanum  
Gigantocotyle anisocotyle  
G. bathycotyle

Bubalus arnee f. bubalis (cont.)

Gastrothylax crumenifer  
G. glandiformis  
G. minutus  
Carmyerius spatiosus  
C. bulbosus  
C. synethes  
Fischoederius elongatus  
F. japonicus

Capra aegargus f. hircus

Paramphistomum liorchis  
Calicophoron calicophorum  
C. cauliorchis  
Orthocoelium scolioceolium  
Gastrothylax crumenifer  
G. glandiformis  
Carmyerius synethes  
Homalogaster paloniae

\*Ovis ammon f. aries

Gastrothylax crumenifer

Cervus unicolor

Calicophoron calicophorum  
C. microbothrioides  
Orthocoelium dinniki  
O. saccocoelium  
Carmyerius synethes  
Fischoederius japonicus

Muntiacus muntjak

Calicophoron calicophorum  
C. microbothrioides  
Orthocoelium scolioceolium  
Homalogaster paloniae  
Carmyerius spatiosus  
Fischoederius elongatus  
F. japonicus

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**SEY, O.: A vietnami gerinces állatok amfisztómumai  
(Trematoda: Amphistomida)**

A szerző vizsgálta a vietnami gerinces állatok amfisztómum élősködőit. Négy fajt halakból, két fajt kétélűtűkből és huszonekét fajt emlősökből mutatott ki. A dolgozatot parazita/gazda és gazda/parazita jegyzék egészíti ki.

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