LARVAL TREMATODES (DIGENEA) OF THE GREAT POND SNAIL, *LYMNAEA STAGNALIS* (L.), (GASTROPODA, PULMONATA) IN CENTRAL EUROPE: A SURVEY OF SPECIES AND KEY TO THEIR IDENTIFICATION

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Summary:

A survey of cercariae and metacercariae (Trematoda, Digenea) from the great pond snail (*Lymnaea stagnalis*) in Central Europe (Austria, Czech Republic, South-East Germany, Poland and Slovak Republic) is presented, based on a study of 3,628 snails examined from 1998 to 2005. A total of 953 (26.3 %) *L. stagnalis* were infected with 24 trematode species comprising 19 species of cercariae and 11 species of metacercariae (six species occurred both as cercarie and metacercarie) of eight families. The dominant cercariae were those of *Opisthioglyphe ranae* (159 hosts infected), *Plagiorchis elegans* (141) (both family Plagiorchiidae) and *Echinoparyphium aconiatum* (153) (Echinostomatidae); 14 double infections were found. The most frequent metacercariae were those of *Neoglyphe locellus* (71) (Omphalometridae), *E. aconiatum* (66), *Echinostoma* sp. (59) and *Moliniella anceps* (48) (Echinostomatidae). In the previous studies carried out in Central Europe, a very similar spectrum of nine trematode families of 22 cercariae determined to species level and 43 types of cercariae reported under generic or provisional names, which can be in many cases conspecific with the previous taxa, were found. A simple key to identification of cercariae and metacercariae, together with their illustrations, is provided.

KEY WORDS : Trematoda, Mollusca, Basommatophora, cercariae, metacercariae, life cycle, prevalence, great pond snail, *Lymnaea stagnalis*.

Résumé : LARVES DE TRÉMATODES (DIGENEA) DE *LYMNAEA STAGNALIS* (L.), (GASTROPODA, PULMONATA) EN EUROPE CENTRALE : ÉTUDE DES ESPÈCES ET CLÉ POUR LEUR IDENTIFICATION

Une étude des cercaires et métacercaires (Trematoda, Digenea) de l'escargot d'eau douce Lymnaea stagnalis *en Europe Centrale (Autriche, République Tchéque, sud-est de l'Allemagne, Pologne et Slovaquie) a porté sur 3628 spécimens prélevés de 1998 à 2005. Un total de 953 (26,3 %)* L. stagnalis *ont été retrouvés infecté par 24 espèces de trematodes, dont 19 espèces de cercaires et 11 de métacercaires (six espèces présentant à la fois des cercaires et des métacercaires) appartenant à huit familles. Les cercaires majoritairement présentes étaient celles d'*Opisthioglyphe ranae *(159 hôtes infestés),* Plagiorchis elegans *(141) (famille des Plagiorchiidae pour les deux) et* Echinoparyphium aconiatum *(153) (Echinostomatidae) ; 14 doubles infestations ont été trouvées. Les métacercaires majoritairement présentes étaient celles de* Neoglyphe locellus *(71) (Omphalometridae),* E. aconiatum *(66),* Echinostoma *sp. (59) et* Moliniella anceps *(48) (Echinostomatidae). Les précédentes études menées en Europe Centrale ont rapporté des résultats proches : neuf familles de trématodes avec 22 cercaires identifiées en tant qu'espèces et 43 types de cercaires répertoriées sous des noms génériques ou provisoires, celles-ci pouvant dans de nombreux cas être conspécifiques avec les précédents taxa. Une clé d'identification des cercaires et des métacercaires avec leurs illustrations est fournie.*

MOTS CLÉS : *Trematode, Mollusque, Basommatophora, cercaire, métacercaire, cycle parasitaire, prévalence, escargot d'eau douce,* Lymnaea stagnalis.

INTRODUCTION

The great pond snail (*Lymnaea stagnalis*) is one of the best known aquatic molluscs with holarctic distribution, occurring in Europe, Asia, North Africa and North America (Jackiewicz, 2000; Beran, 2002; Glöer, 2002). In Europe, it is widely distributed and lives even in some parts of the Baltic sea with low salinity (Jackiewicz, 2000). It can be found predominantly in lower altitudes (150-250 m) where it inhabits slowly flowing or still waters, ponds, blind arms, pools and also periodic swamps, and it is able to survive short dry periods (Jackiewicz, 2000; Beran, 2002).

Being the most common and conspicuous snail, together with *Planorbarius corneus* (Planorbidae), *L. stagnalis* was examined in the very first larval trematode investigations of Nitzsch (1817), La Valette (1855), Pagenstecher (1857) and later Lühe (1909). Until today, *L. stagnalis* is still of interest, being the intermediate host not only of a wide variety of larval trematodes, which are among the most common species (*Echinoparyphium aconiatum*, *Opisthioglyphe ranae*, *Diplostomum pseudospathaceum*) in Central Europe (Faltýnková & Haas, 2006), but also of the causative agents of cercarial dermatitis – cercariae of bird schistosomes (genus *Trichobilharzia*), which are studied quite intensively in Austria, Czech Republic, Germany and Poland (Allgöwer, 1990; Kolářová *et al* 1997; Kolářová &

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Horák, 1996; Konecny *et al.*, 1999; Dvorák *et al.*, 1999; Kalbe *et al.*, 2000; Żbikowska, 2003, 2004; Sattmann *et al.*, 2004; Horák & Kolářová, 2005; Rudolfová *et al.*, 2005; Hertel *et al.*, 2006).

Loy & Haas (2001) found 18 species of trematode larvae during their long-time investigation into the behaviour of cercariae from *L. stagnalis* in Germany. They found out that the spectrum did not change over the period of 20 years. The species composition was also similar to reports from the 19th century (see references above). However, a substantial part of the cercariae found by Loy & Haas (2001) were not determined to species. Cercariae with invalid or provisional names were often listed in articles dealing with the trematode fauna of *L. stagnalis* published in hardly accessible journals of regional importance. In some countries only few records of cercariae other than bird schistosomes exist (Konecny *et al.*, 1999). Several attempts to compile an atlas of cercariae (Combes, 1980) or keys to identification of different types of cercariae (Odening, 1962a; Palm, 1966a, Blair, 1977) were made, but these are of difficult use. And recently, there is no updated and comprehensive information about larval trematodes of the great pond snail.

Because of the large size of the great pond snail, producing large amounts of cercariae over a long period, it is a favoured model organism for studies on parasite-host physiology, immunology and behaviour. However, correct identification of larval trematodes is the premise for any studies and also the knowledge of species composition is important for studies on the epidemiology of veterinary or medically important trematodes and for ecology of trematode communities. Therefore, a survey of larval trematodes in the great pond snail in Central Europe, based on original data from five countries and revised records from literature, is presented here, together with a simple key to identification suitable also for non-trematodologists and illustrations of the most common species. Detailed morphological descriptions of larval trematodes with more information about their distribution and life cycles will be presented in a separate monograph.

MATERIAL AND METHODS

Sampling and handling of snails

Snails of *Lymnaea stagnalis* were sampled from April 1998 to October 2005 in still and slightly flowing water bodies of drainage areas of major rivers of Central Europe (Danube, Elbe, Main, Odra, Tisa). The regions studied comprised pond systems of South Bohemia and Moravia in the Czech Republic (see Faltýnková, 2005), in South-East Germany (see Faltýnková & Haas, 2006), in the reserve of Milicz in Poland and ponds near Danube in Germany; blind arms of Danube in Austria and the Slovak Republic and channels along the Tisa River in East Slovakia (Fig. 1).

Molluscs (n = 3,628) were sampled with a strainer or hand picked on shores. In the laboratory, they were separated into glass containers with tap water (100 ml). Shedding of cercariae was stimulated by light and heat for 4-6 hours. The internal morphology of cercariae and metacercariae was documented by drawings

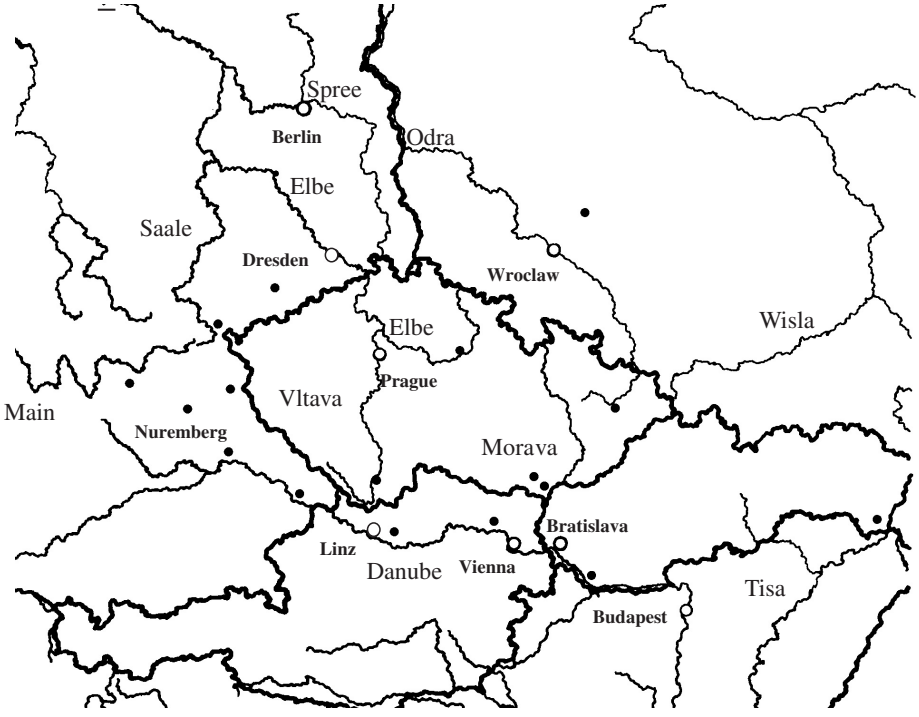

Fig. 1. – Survey of localities (●) where samples of snails were taken.

with aid of a drawing tube. Neutral red (to observe paraoesophageal glands of echinostomatids) and Nile blue were used for vital staining; urine solution was used to study the excretory system. For measurements, cercariae were fixed in hot 4 % formaldehyde solution. Afterwards, a part of the molluscs were dissected to detect larval stages other than cercariae. Cercariae were identified with help of publications of Combes (1980), Niewiadomska (1986), Našincová (1992) and Niewiadomska *et al.* (1997). The system proposed by Gibson *et al.* (2002), Jones *et al.* (2005) and Bray *et al.* (2007) is followed.

SURVEY OF LITERARY DATA

To provide a complete survey of larval trematodes from *Lymnaea stagnalis*, literary data from Austria, Czech and Slovak Republics, Germany, Hungary and Poland were compiled to supplement the original data obtained during the present survey and in recent records of Faltýnková (2005) and Faltýnková & Haas (2006). Records based on experimental infections were not considered. For better orientation, valid names of adult trematodes were provided together with original invalid or provisional names appearing in the literature.

A simple key to species of cercariae and metacercariae from *L. stagnalis* (Table II) is presented. Larval stages not identified at least to genus (Table III) are not included in the key, except for *Cercaria spinulosa* Ginetsinskaya, 1959. Only illustrations of common species recorded in the present study are presented; further figures can be found in publications of authors listed in Tables II and III.

RESULTS

In the present survey, a total of 26.3 % *Lymnaea stagnalis* were infected (Table I) with 24 trematode species comprising 19 species of cercariae and 11 species of metacercariae (six species occurred both as cercarie and metacercarie) of eight families (Table II). The dominant cercariae were those of *Opisthioglyphe ranae* (159 hosts infected), *Plagiorchis elegans* (141) (both Plagiorchiidae) and *Echinoparyphium aconiatum* (153) (Echinostomatidae). The most frequent metacercariae were those of *Neoglyphe locellus* (71) (Omphalometridae), *E. aconiatum* (66), *Echinostoma* sp. (59) and *Moliniella anceps* (48) (Echinostomatidae). Metacercariae of *O. ranae* (3), *P. elegans* (1) and strigeids (1) were found less frequently.

In the previous studies carried out in Central Europe, as many as 22 cercariae determined to species level and 43 taxa recorded under generic or provisional names of nine families were reported from *L. stagnalis* (Tables II, III).

KEY TO CERCARIAE

1 Cercaria without tail (cercariaeum); intestinal caeca form cyclocoel ..... *Cyclocoelum microstomum*

Cercaria with tail; intestinal caeca blind..... 2

2 Tail long, simple 3

Tail forked (furcocercariae) 17

3 Cercaria with two suckers, ventral sucker more or less equatorial 4

Monostomous cercaria .... *Notocotylus attenuatus* (Fig. 5A)

4 Collar and collar spines present; excretory granules (between oral and ventral sucker); excretory vesicle more or less spherical 5

Collar and collar spines and excretory granules absent; stylet on dorsal part of oral sucker; excretory vesicle Y-shaped 12

5 Dorsal collar spines arranged in one row 6

Dorsal collar spines in two rows; number of collar spines different 7

6 Cercaria large (body 400-500 µm long); excretory canals form finger-like processes anteriorly; excretory granules conspicuous; 47-50 collar spines .... *Cathaemasia hians* (Fig. 3C-E)

Cercaria smaller; excretory canals form simple loops anteriorly; excretory granules small; 46-55 collar spines ..... *Hypoderaeum conoideum* (Fig. 4B, F)

7 Tail with fin folds; paraoesophageal glands may be present (stain intensively with neutral red) 8

Tail without fin folds; no paraoesophageal glands....9

Country	Examined	Infected	%	No. species	
				Cercariae	Metacercariae
Austria	203	22	10.8	3	–
Czech Republic	1,078	409	37.9	14	7
Germany	1,219	187	15.3	10	–
Poland	669	273	40.8	13	2
Slovak Republic	459	62	13.5	7	5
Total	**3,628**	**953**	**26.3**	**19**	**11**

Table I. – Survey of *Lymnaea stagnalis* examined.

Valid name	Distribution	Original names and authorities
Cyclocoelidae Stossich, 1902		
Cyclocoelum microstomum (Creplin, 1829)	CZ**	Žďárská (1963)
Diplostomidae Poirier, 1886		
**Diplostomum pseudospathaceum* Niewiadomska, 1984	CZ, D, PL	*Diplostomum spathaceum* of Wisniewski (1958), Styczyńska-Jurewicz (1959), Žďárská (1963, 1964a), Odening (1964a) in part, Odening (1965a), Palm (1966a), Gottschalk (1971), Vojtek & Vojtková (1971), Fleischerová (1972), Gelnar (1980); Furcocercaria sp. 1 of Odening (1962a); Cercaria sp. 1 of Bursian-Hartung (1965) in part; Niewiadomska (1986), Našincová (1992), Loy & Haas (2001), Niewiadomska & Laskowski (2002), Faltýnková (2005), Faltýnková & Haas (2006), present study
Tylodelphys clavata (Nordmann, 1832)	CZ	Našincová (1992)
Strigeidae Railliet, 1919		
**Australapatemon burti* (Miller, 1923)	CZ, D, PL	*Apatemon burti* (Miller, 1923) of Zajíček (1971), Našincová (1992); *Apatemon gracilis* of Zajíček & Valenta (1964); *Apatemon gracilis gracilis* of Vojtek & Vojtková (1971); *Apatemon minor* of Loy & Haas (2001); Faltýnková & Haas (2006), present results
Australapatemon minor (Yamaguti, 1933)	CZ	*Apatemon minor* of Zajíček (1971), Našincová (1992)
Cotylurus brevis Dubois et Rausch, 1950	CZ, D	Odening (1965a), Zajíček (1971)
Cotylurus cornutus (Rudolphi, 1808)	CZ, D	Žďárská (1963, 1964a), Odening (1965a), Zajíček (1971)
Schistosomatidae Stiles & Hassall, 1898		
**Trichobilharzia szidati* Neuhaus, 1952	A, CZ, D, PL, SK	*Cercaria ocellata* La Valette, 1855 of Lühe (1909), Wisniewski (1958), Bertman & Wojciechowska (1974), Farkaš (1976); *Trichobilharzia ocellata* of Emmel (1947), Žďárská (1963), Zajíček & Valenta (1964) in part, Odening (1965a), Palm (1966a), Gelnar (1980), Našincová (1992) in part, Loy & Haas (2001); *Cercaria Trichobilharziae ocellatae* of Bursian-Hartung (1965); *Cercaria Trichobilharziae szidati* of Dönges (1965); *Trichobilharzia* sp. König (2001); Neuhaus (1952), Kolářová *et al.* (1992) in part, Pilz *et al.* (1995), Kolářová & Horák (1996) in part, Kolářová *et al.* (1997) in part, Dvořák *et al.* (1999), Sattmann *et al.* (2004), Rudolfová *et al.* (2005), Faltýnková (2005), Faltýnková & Haas (2006); present study
Sanguinicolidae von Graff, 1907		
Sanguinicola inermis Plehn, 1905	CZ, D	*Cercaria cristata* La Valette, 1855 of Lühe (1909), Wisniewski (1958); *Sanguinicola* sp. (*inermis*) of Gelnar (1980); Scheuring (1922), Odening (1965a)
Echinostomatidae Looss, 1899		
**Echinoparyphium aconiatum* Dietz, 1909	CZ, D, PL, SK	*Cercaria echinata* Siebold, 1837 of La Valette (1855), Lühe (1909), Zajíček (1963), Bertman & Wojciechowska (1974), Bertman (1980); *Pseudechinoparyphium echinatum* Kanev & Vasilev, 1986 of Loy & Haas (2001); Žďárská (1964a), Odening (1965a, 1965b), Gelnar (1980), Našincová (1992), Faltýnková (2005), Faltýnková & Haas (2006), present study
**Echinoparyphium recurvatum* (Linstow, 1873)	CZ, D, PL	Wisniewski (1958), Žďárská (1963, 1964a) in part, Odening (1965a, 1965b), Nezvalová (1970), Balúsek & Vojtek (1973) in part, Gelnar (1980); Faltýnková (2005), Faltýnková & Haas (2006), present study
**Echinostoma revolutum* (Fröhlich, 1802)	A, CZ, D, PL	*Cercaria echinostomi-revoluti* of Zajíček (1963); *Cercaria* sp. 10 of Bursian-Hartung (1965); Wisniewski (1958), Žďárská (1963, 1964a) in part, Odening (1965a) in part, Gelnar (1980), Našincová (1992), Nezvalová (1970) in part, Kanev *et al.* (1993), Loy & Haas (2001), Faltýnková (2005), Faltýnková & Haas (2006), present study
**Hypoderaeum conoideum* (Bloch, 1782)	CZ, D, PL, SK	*Cercaria affinis* Wesenberg-Lund, 1934 of Ahmed (1959); Odening (1965a), Nezvalová (1970), Balúsek & Vojtek (1973), Gelnar (1980), Našincová (1992), Loy & Haas (2001), Faltýnková & Haas (2006), present study
**Isthmiophora melis* (Schrank, 1788)	CZ, D, PL	*Euparyphium melis* of Žďárská (1964b); Grabda-Kazubska & Laskowski (1996), Loy & Haas (2001), present study
**Moliniella anceps* (Molin, 1858)	CZ, D	Odening (1964b), Faltýnková (2005)
**Paryphostomum radiatum* (Dujardin, 1845)	CZ, PL	present study
Cathaemasiidae Fuhrmann, 1928		
Cathaemasia hians (Rudolphi, 1809)	CZ, PL	Cercaria sp. 2 of Balúsek & Vojtek (1973); Grabda-Kazubska *et al.* (1990)
Notocotylidae Lühe, 1909		
**Notocotylus attenuatus* (Rudolphi, 1809)	A, CZ, D, PL	*Cercaria notocotyli-attenuati* of Zajíček (1963); *Cercaria monostomi* Linstow, 1884 of Bursian-Hartung (1965), Bertman & Wojciechowska (1974), Bertman (1980); *Notocotylus* sp. of Konecny *et al.* (1999); Žďárská (1963), Nezvalová (1970) in part, Balúsek & Vojtek (1973), Gelnar (1980), Našincová (1992), Faltýnková (2005), present study

Table II *(to be continued).*

Valid name	Distribution	Original names and authorities
Plagiorchiidae Lühe, 1901		
**Haematoloechus similis* (Looss, 1899)	CZ	present study
**Opisthioglyphe ranae* (Fröhlich, 1791)	A, CZ, D, PL, SK	*Cercaria armata* of Pagenstecher (1857); *Cercaria armata* Siebold of La Valette (1855), Lühe (1909); *Xiphidiocercaria* sp. 2 of Odening (1962b); *Cercaria opisthioglyphe-ranae* of Zajíček (1963); *Cercaria* sp. 7 of Bursian-Hartung (1965); Styczyńska-Jurewicz (1961), Žďárská (1963, 1964a) in part, Palm (1966b), Grabda-Kazubska (1969), Nezvalová (1970), Gelnar (1980), Dimitrov *et al.* (1989), Našincová (1992), Faltýnková (2005), Faltýnková & Haas (2006), present study
**Plagiorchis elegans* (Rudolphi, 1802)	A, CZ, D, PL SK	*Cercaria* sp. 6 of Bursian-Hartung (1965); Styczyńska-Jurewicz (1961), Našincová (1992), Faltýnková (2005), Faltýnková & Haas (2006), present study
Plagiorchis laricola Skryabin, 1924	CZ	Žďárská (1966), Samnaliev *et al.* (1983), Gelnar (1980)
**Plagiorchis maculosus* (Rudolphi, 1802)	CZ	Bušta (1987), Našincová (1992), Faltýnková (2005)
Omphalometridae Odening, 1960		
**Neoglyphe sobolevi* Shaldybin, 1953	CZ, D, PL	Našincová *et al.* (1989), Našincová (1992), present study

* Species found in recent studies.
**A – Austria, CZ – Czech Republic, D – Germany, PL – Poland, SK – Slovak Republic.

Table II. – Survey of species of cercariae reported from *Lymnaea stagnalis* in Europe.

8 Cercaria large (500-700 µm), with one large dorso-ventral fin fold; tip of tail without contractile process; 35 collar spines; paraoesophageal glands absent .... *Moliniella anceps* (Fig. 4C, G)

Cercaria small (250-300 µm); with seven small, inconspicuous fin folds; tip of tail forms contractile process; 37 collar spines; 12 paraoesophageal glands ..... *Echinostoma revolutum* (Fig. 4A, E)

9 Cercaria with numerous small excretory granules; 37 or more collar spines 11

Few large excretory granules; 27 collar spines 10

10 Oral and ventral sucker possess crista; body smooth ..... *Paryphostomum radiatum* (Fig. 4D, H)

Suckers without cristae; spinose body .... *Isthmiophora melis*

11 Cercaria large (500-600 µm); 37 collar spines; angle spines larger .... *Echinoparyphium aconiatum* (Fig. 3F, G)

Cercaria small (200-250 µm); 45 collar spines of same length .... *Echinoparyphium recurvatum* (Fig. 3H)

12 Cercaria small (100-120 µm); stylet straight, without anterior or posterior thickenings; penetration glands pre- and postacetabular .... *Haematoloechus similis* (Fig. 5B, C)

Cercariae larger; stylet with anterior thickening (see Fig. 5E, G); penetration glands preacetabular 13

13 Cercaria large (270-320 µm), dark; stylet small, with incomplete basis and small anterior thickening; suckers of about same size ..... *Opisthioglyphe ranae* (Fig. 5D, E)

Cercaria small (130-200 µm); stylet conspicuous, with complete basis and larger anterior dilatation; oral sucker larger than ventral sucker 14

14 Seven pairs of penetration glands 16

Seven penetration glands on one side and eight on the other 15

15 Stylet without thickening at its basis..... ..... *Plagiorchis elegans* (Fig. 5F, G)

Stylet with thickening at its basis; posterior penetration glands paraacetabular ... *Neoglyphe sobolevi* (Fig. 5H, I)

16 Fat inclusions in body parenchyma; stylet without column ..... *Plagiorchis maculosus*

No fat inlusions in body; stylet with column .... *Plagiorchis laricola*

17 Body with crista; pharynx absent (lophocercous cercaria) ..... *Sanguinicola inermis*

Body without crista; pharynx present or within anterior organ 18

18 Eye-spots with black pigment; five pairs of large penetration glands; furcae with fins .... *Trichobilharzia szidati* (Fig. 3A, B)

Eye-spots colourless; different number of penetration glands; furcae without fins 19

19 Two pairs of preacetabular penetration glands 20

Penetration glands postacetabular 23

20 One row of hooked conspicuous spines on ventral sucker; flame cell formula 2 [(2 + 2) + (2 + (2))] = 16 ... . *Tylodelphys clavata* (Fig. 2G, H)

Three and more rows of small spines on ventral sucker; flame cell formula 2 [(2 + 2) + (2 + 2 + (2))] = 20 21

21 Cercaria with conspicuous large spines on whole body ..... *Cercaria spinulosa*

Body spinose except for oval area around ventral sucker 22

22 Cercaria with long oesophagus (longer than pharynx) ..... *Cotylurus cornutus* (Fig. 2I)

Oesophagus short (as long as pharynx) .... *Cotylurus brevis*

Family/Generic name	Distribution	Original names and authorities
Diplostomidae Poirier, 1886		
Diplostomum sp.	CZ	*Diplostomum* sp. (= *Cercaria chromatophora* Brown, 1931) of Zajíček & Valenta (1964)
Strigeidae Railliet, 1919		
?*Australapatemon*	D	Cercaria furcata Nitzsch, 1807 of Nitzsch (1817), La Valette (1855), Lühe (1909)
Cotylurus sp.	CZ	*Cotylurus erraticus* (Rudophi, 1809) of Zajíček & Valenta (1964), Vojtek & Vojtková (1971)
Cotylurus sp.	CZ	*Cotylurus* sp. II Ginetsinskaya, 1959 of Žďárská (1963), Opravilová (1969), Gelnar (1980)
?*Ichthyocotylurus* sp.	CZ	*Cercaria spinulosa* Ginetsinskaya, 1959 of Zajíček & Valenta (1964)
Schistosomatidae Stiles & Hassal, 1898		
Trichobilharzia sp.	CZ	Rudolfová *et al.* (2005)
Sanguinicolidae von Graff, 1907		
Sanguinicola spp.	A	Konecny *et al.* (1999)
Echinostomatidae Looss, 1899		
Echinoparyphium ?recurvatum	CZ	*Cercaria echinoparyphii* sp. 1 of Zajíček (1963), Nezvalová (1970)
?*Isthmiophora melis/?Paryphostomum* sp.	CZ	*Paryphostomum* sp. 2 of Našincová (1992)
Isthmiophora/Paryphostomum	D	*Cercaria fallax* Pagenstecher, 1857 of Pagenstecher (1857), Lühe (1909), Bursian-Hartung (1965)
?*Echinoparyphium aconiatum*	CZ	*Cercaria helvetica* XXII Dubois 1929 of Nezvalová (1970); *Echinostoma paraulum* Dietz, 1909 of Zajíček (1963), Nezvalová (1970), Gelnar (1980); *Cercaria* sp. 1 of Balŭsek & Vojtek (1973)
	D	*Echinostome cercaria* with 29 collar spines of Odening (1965b)
Notocotylidae Lühe, 1909	CZ	*Notocotylus* sp. Ginetsinskaya 1959 of Nezvalová (1970) in part
Plagiorchiidae Lühe, 1901		
?*Leptophallus*	CZ	*Cercaria helvetica* VI Dubois, 1927 of Zajíček (1963)
?*Leptophallus*	D	*Cercaria secunda* Sinitzin, 1905 of Lühe (1909)
*cf. *Leptophallus* sp.	PL	present study
**Plagiorchis* sp. 1	D, SK	present study
**Plagiorchis* sp. 2	SK	present study
?*Plagiorchi*s sp.	CZ	*Cercaria helvetica* IV Dubois 1927 of Zajíček (1963)
	CZ	*Cercaria helvetica* XXVII Dubois 1929 of Žďárská (1963)
	CZ	*Cercaria limnaeae-ovatae* Linstow 1884 of Žďárská (1963)
	D	*Cercaria tenuispina* of Lühe (1909)
	CZ	*Xiphidiocercaria* sp. 1 of Balŭsek & Vojtek (1973), Gelnar (1980)
	CZ	*Xiphidiocercaria* sp. 3 Balŭsek & Vojtek, 1973 of Gelnar (1980)
	D	*Xiphidiocercaria* sp. 3 of Odening (1962b)
	D	*Xiphidiocercaria* sp. 4 of Odening (1962b)
	D	*Xiphidiocercaria* sp. 5 of Odening (1962b)
	CZ, D	*Xiphidiocercaria* sp. A Palm, 1966 of Nezvalová (1970); Gelnar (1980)
	D	*Xiphidiocercaria* sp. C Palm, 1966 of Palm (1966b)
		Xiphidiocercaria I Ginetsinskaya 1959 of Palm (1966b), Nezvalová (1970), Gelnar (1980)
	CZ	*Xiphidiocercaria* II Ginetsinskaya 1959 of Žďárská (1963)
	CZ, D	*Xiphidiocercaria* sp. III Ginetsinskaya 1959 of Nezvalová (1970); Palm (1966b); Gelnar (1980)
	CZ, D	*Xiphidiocercaria* sp. VI Ginetsinskaya, 1959 of Palm (1966b); Nezvalová (1970); Gelnar (1980)
Lissorchiidae Poche, 1926	D	*Asymphylodora* sp. of Odening (1965a)
?Lissorchiidae	D	*Cercariaeum* sp. A of Palm (1967)
Species incertae sedis	A, D	Unidentified echinostomous cercariae of Konecny *et al.* (1999), Loy & Haas (2001)
	A, D	Unidentified furcocercariae of Konecny *et al.* (1999), Loy & Haas (2001)
	A, D	Unidentified xiphidiocercariae of Konecny et al. (1999), Loy & Haas (2001)
misidentified	PL	*Cercaria auriculariae* Zdun of Bertman & Wojciechowska (1974)
Strigeidae?	D	*Cercaria brunnea* Diesing, 1850 of Lühe (1909)
furcocercaria	D	*Cercaria fissicauda* La Valette, 1855 of Lühe (1909)
misidentified	CZ	*Cercaria gracilis* Wesenberg-Lund 1934 of Zajíček (1963)
misidentified	PL	*Cercaria longiremis* Wesenberg-Lund, 1934 of Bertman (1980)
misidentified	PL	*Cercaria onusta* Zdun of Bertman & Wojciechowska (1974)
misidentified	PL	*Cercaria* Petersen I Petersen of Bertman & Wojciechowska (1974)
misidentified	PL	*Cercaria pseudogracilis* Zdun of Bertman & Wojciechowska (1974), Bertman (1980)
misidentified	CZ, D	*Xiphidiocercaria* sp.VII Odening 1962 of Nezvalová (1970) in part

Table III – Survey of species of cercariae identifited to genus or with provisional names reported from *Lymnaea stagnalis* in Europe.

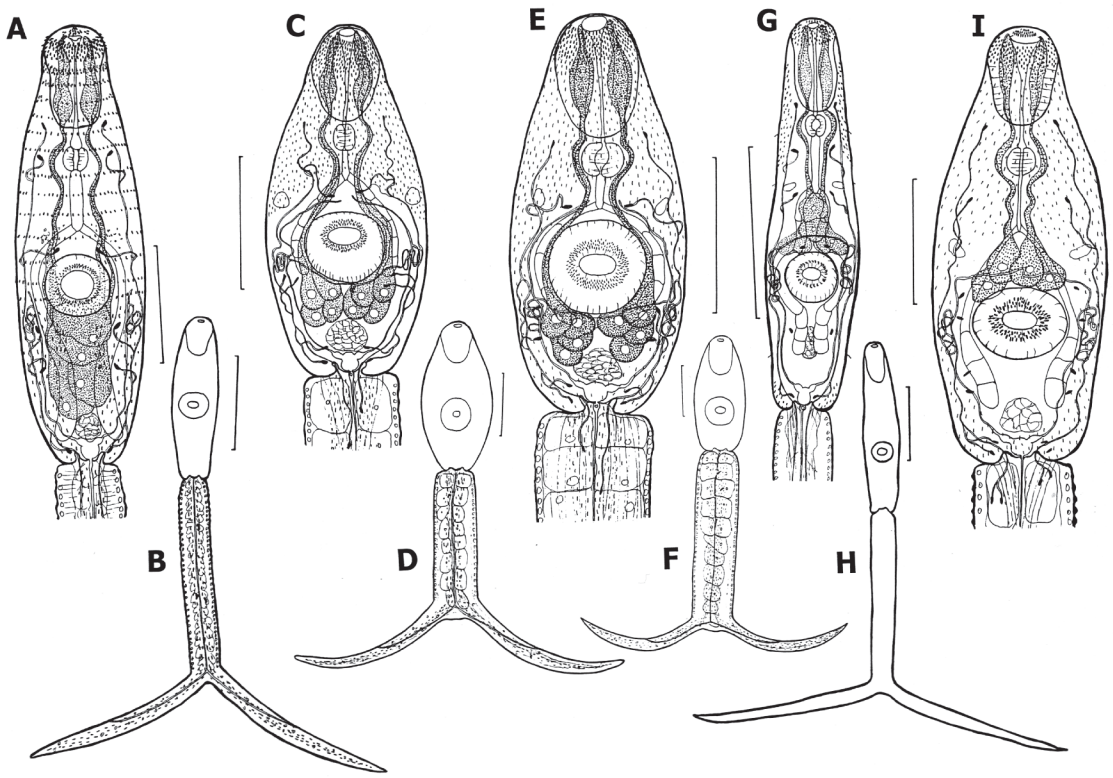

Fig. 2. – *Diplostomum pseudospathaceum* (A, B); *Australapatemon burti* (C, D); *Australapatemon minor* (E, F); *Tylodelphys clavata* (G, H); *Cotylurus cornutus* (I); A, C, E, G, I – detail of body, scale bars: A, C, E, I – 50 µm, G – 100 µm; B, D, F, H – whole cercaria, scale bars: B, H – 100 µm, D, F – 50 µm.

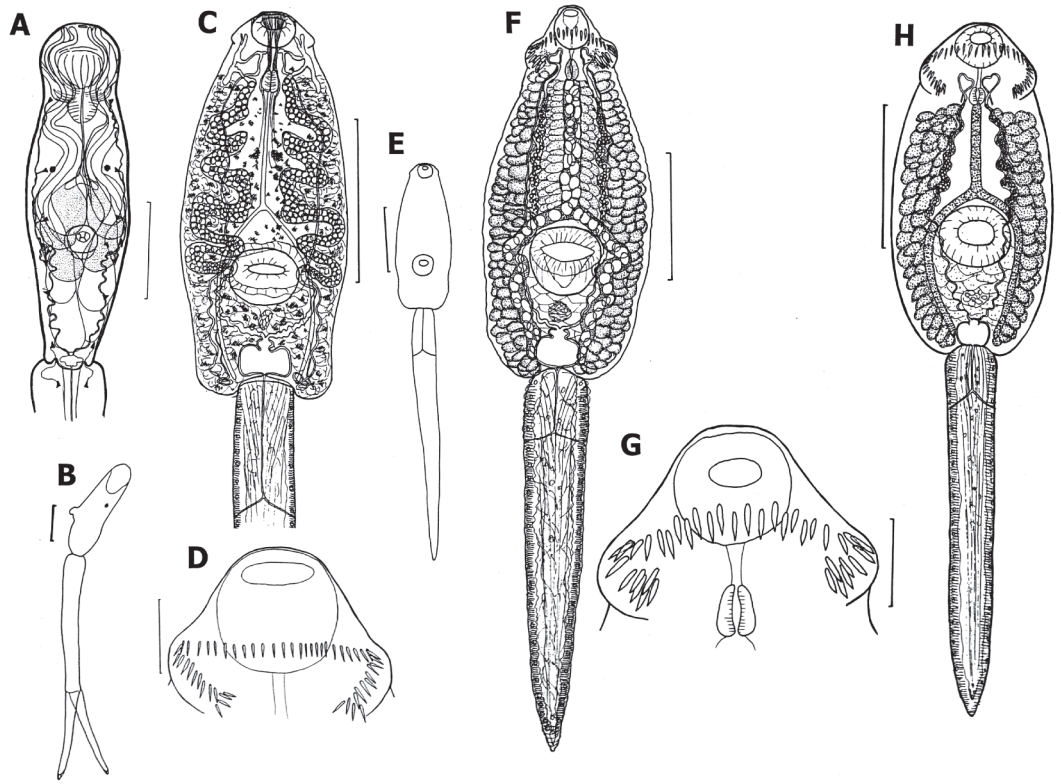

Fig. 3. – *Trichobilharzia szidati* (A, B); *Cathaemasia hians* (C, D, E); *Echinoparyphium aconiatum* (F, G); *Echinoparyphium recurvatum* (H); A, C – detail of body, scale bars: A – 100 µm, C – 200 µm; B, E, F, H – whole cercaria, scale bars: B, H – 100 µm, E, F – 200 µm; G, D – detail of anterior part with collar spines, scale bar: 50 µm.

23 Two pairs of large postacetabular penetration glands; anterior organ with large conspicuous spines; caeca long, reach to body end; flame cell formula 2 [(1 + 1 + 1) + (1 + 1 + 1) + (2)] = 16 .... *Diplostomum pseudospathaceum* (Fig. 2A, B)

Four pairs of small, poorly visible postacetabular penetration glands; anterior organ with fine spines; caeca short, reach to posterior end of ventral sucker; flame cell formula 2 [2 + (2 + 2 + (1))] = 14 24

24 Body spined only on its anterior part .... *Australapatemon burti* (Fig. 2C, D)

Whole body spinose ..... *Australapatemon minor* (Fig. 2E, F)

KEY TO METACERCARIAE

1 Metacercaria spherical 2

Metacercaria oval 8

2 Cyst thick-walled; excretory granules and collar spines present; encysted in pericardium, kidney, less often in musculature 3

Cyst thin-walled; collar spines absent 6

3 Metacercaria large (about 270-380 µm in diameter) 4

Metacercaria small (about 140 mm in diameter) 5

4 Collar of excysted metacercaria narrower than long body, with 35 spines of same length; three spherical anlagen of reproductive organs .... *Moliniella anceps* (Fig. 6D)

Collar of excysted metacercaria of same width as short body; 37 collar spines, with conspicuous, larger angle spines .... *Echinoparyphium aconiatum* (Fig. 6A)

5 Cercaria with 37 collar spines; excretory granules larger ..... *Echinostoma* sp. (Fig. 6C)

Cercaria with 45 collar spines; excretory granules smaller ..... *Echinoparyphium recurvatum* (Fig. 6B)

6 Refractile granules in Y-shaped excretory vesicle; released stylet floating in cyst; anlagen of reproductive organs absent; encysted mainly in musculature 7

Refractile granules and stylet absent; anlagen of reproductive organs present ..... *Asymphylodora* sp.

7 Metacercaria large (200 µm), shape of stylet – see Fig. 5E ..... *Opisthioglyphe ranae* (Fig. 6E)

Metacercaria small (130 µm); shape of stylet – see Fig. 5G ..... *Plagiorchis* sp. (Fig. 6F)

8 Cyst thin-walled; encysted in musculature (edge of mantle); Y-shaped transparent excretory vesicle .... *Neoglyphe locellus* (Kossack, 1910) (Fig. 6G)

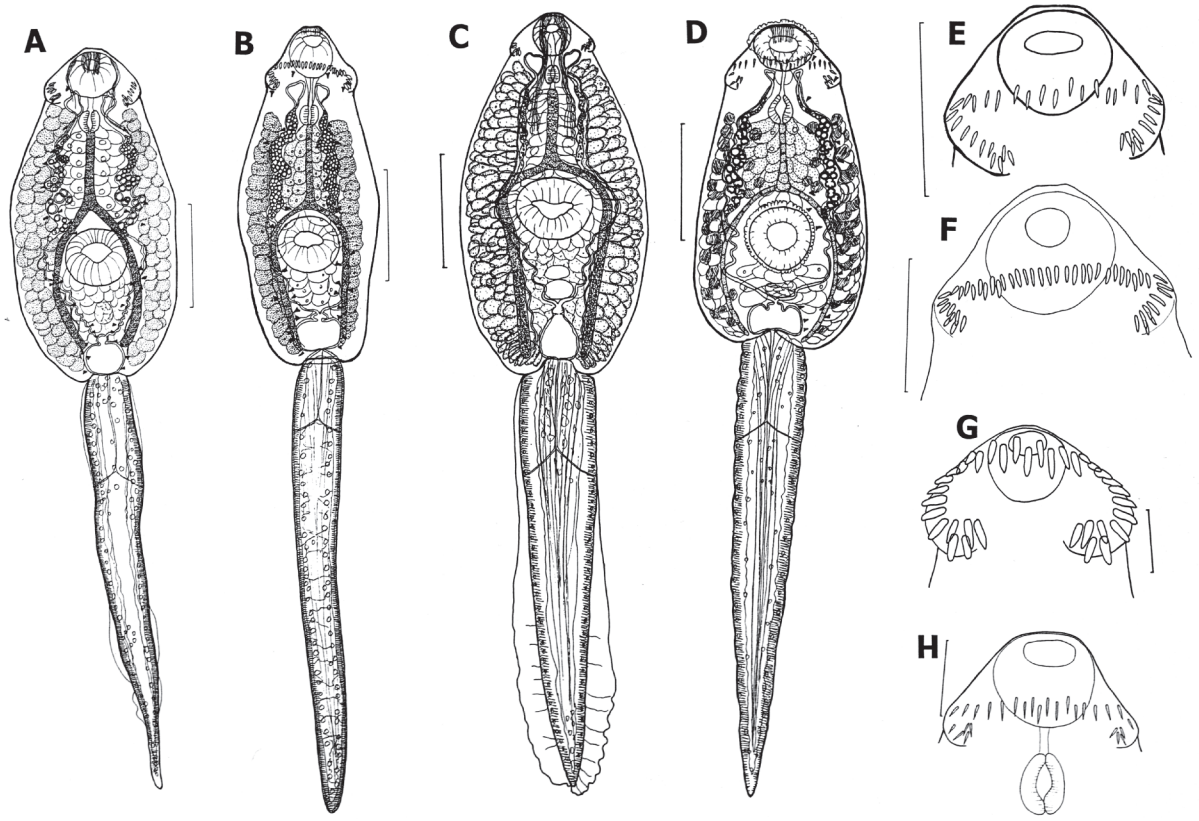

Fig. 4. – *Echinostoma revolutum* (A, E); *Hypoderaeum conoideum* (B, F); *Moliniella anceps* (C, G); *Paryphostomum radiatum* (D, H); A-D – whole cercaria, scale bars: A, B, D – 100 µm, C – 200 µm; E-H – detail of anterior part with collar spines, scale bar: 50 µm.

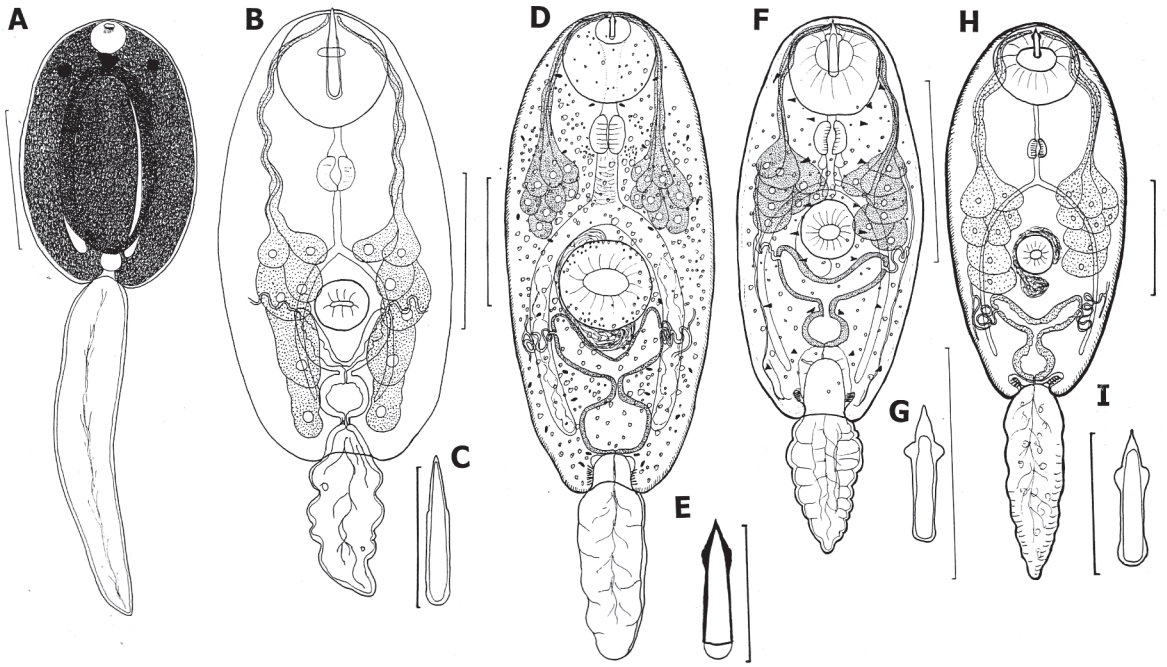

Fig. 5. – *Notocotylus attenuatus* (A); *Haematoloechus similis* (B, C); *Opisthioglyphe ranae* (D, E); *Plagiorchis elegans* (F, G); *Neoglyphe sobolevi* (H, I); A, B, D, F, H – whole cercaria, scale bars: A – 200 µm, B – 50 µm, D, F, H – 100 µm; C, E, G, I – stylet, scale bars: C, E, I – 30 µm, G – 50 µm.

DISCUSSION

In *Lymnaea stagnalis* a total of 24 trematode species comprising 19 species of cercariae and 11 species of metacercariae of eight families were found recently. A rather similar fauna of 22 cercariae determined to species level and 43 cercariae reported under generic or provisional names of nine trematode families were found in former studies carried out in Central Europe.

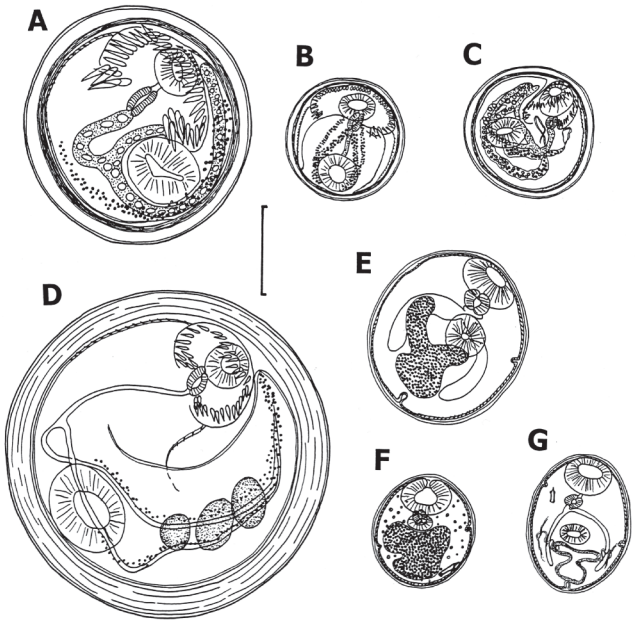

Fig. 6. – *Echinoparyphium aconiatum* (A), *Echinoparyphium recurvatum* (B), *Echinostoma* sp. (C), *Moliniella anceps* (D), *Opisthioglyphe ranae* (E), *Plagiorchis* sp. (F), *Neoglyphe locellus* (G); scale bar 100 µm.

However, it must be assumed that the real number of existing species is almost certainly much lower than the latter number of 43 taxa, because some species are most probably conspecific with the determined ones. Especially xiphidiocercariae (Plagiorchiidae) with quite similar morphological characteristics were reported under several provisional names; in Table III trematodes with insufficient, dubious or missing descriptions were included. Unfortunately, the real identity of these records cannot be worked out because of insufficient or lacking descriptions. In the group of *Species incertae sedis*, mainly cercariae obviously misidentified without any descriptions were included, because they were assigned to species being specific only for planorbid snails that never occur in lymnaeids.

The dominant cercariae were those of *Opisthioglyphe ranae*, *Echinoparyphium aconiatum* and *Plagiorchis elegans* of the families Plagiorchiidae and Echinostomatidae. These three species are widely distributed in Europe and were found as the most frequent in all countries where samples were undertaken.

The most frequent metacercariae were those of *Neoglyphe locellus* (Omphalometridae), with cercariae being most abundant in *Planorbarius corneus* (Planorbidae). The second most common were metacercariae of the

family Echinostomadidae: *E. aconiatum*, *Echinostoma* sp. and *Moliniella anceps*, which often occurred together. This family contains most species encysting in molluscs. Metacercariae of plagiorchids were found less frequently, since *Plagiorchis elegans* is reported to use snails accidentally. However, the common *Opisthioglyphe* ranae is known to encyst in molluscs. The strigeid metacercariae could belong to a species of *Cotylurus*, which encyst frequently in molluscs (Našincová, 1992).

The highest species diversity of cercariae in *L. stagnalis* was found in the pond systems of the Czech Republic and Poland. The number of species found in Germany was slightly lower, but the spectrum of cercariae (18 species) in the same region found by Loy & Haas (2001), was not much higher. In Austria and the Slovak Republic more blind arms of the Danube river than ponds were examined and lower numbers of *L. stagnalis* could be sampled.

Only one species of the family Strigeidae, *Australapatemon burti*, was found in the present study. The genus *Australapatemon*, established by Sudarikov (1959), was regarded as a subgenus of *Apatemon* by Dubois (1968). For a long time the species of *Australapatemon* were reported as *Apatemon gracilis* (as listed in Table II), although the cercariae of *Australapatemon* differ in morphology and biology from those of *Apatemon* (long vs. rudimentary caeca, different flame cell formula and body spination, metacercariae in leeches vs. fishes, Našincová 1992). No cercariae of *Cotylurus* were found in this study, although several species were recorded mainly by the Czech authors (Tables II, III). *Cotylurus erraticus* (Rudolphi, 1819) (transferred to *Ichthyocotylurus*) was erroneously identified by Zajíček & Valenta (1964). Their species actually belongs to the genus *Cotylurus* and probably represents a separate species (Table III). It could not be assigned to any other known species of *Cotylurus* because of a different type of body spination (spination the same as in *C. cornutus* except for an aspinose part in the area of the excretory vesicle).

Schistosome cercariae occurring in *L. stagnalis* were identified as *Trichobilharzia szidati*, although Rudolfová *et al.* (2005) found another cryptic species in this snail host (listed in Table III). The prevalence of *T. szidati* was very similar in most countries (Czech Republic, Germany and Poland). It was not found in Austria, presumably because of a short sampling period. Nearly the same species composition of echinostomes as in previous surveys was found in the present study. Cercariae with cristae on suckers were considered as *Paryphostomum radiatum* and those without cristae could be *Isthmiophora melis* (Table II). Grabda-Kazubska & Laskowski (1996) redescribed the life cycle of *I. melis* and found that it is difficult to separate these two species, even with the aid of chaetotaxy. However, *P. radiatum* has a crista on the oral and ventral sucker (Našincová *et al.*, 1993), which could be considered as a distinguishing feature, and the species characterized by Našincová (1992) as *Paryphostomum* sp. without cristae could be *I. melis*. Grabda-Kazubska & Laskowski (1996) noted that *Cercaria fallax* Pagenstecher, 1857 was very probably *I. melis*.

The spectrum of plagiorchiid species was also very similar as in former surveys, except for *Haematoloechus similis*, reported only from planorbid snails. The species of *Plagiorchis elegans* and *P. laricola* were both included in Table II, although it is still unclear if they are separate species or not (Našincová, 1992). Since there exist features to distinguish them, they were also included in the key, although the characters (number of penetration glands, column in stylet) are difficult to observe.

It can be assumed that the trematodes from *L. stagnalis* are rather well known and their distribution exceeds the area of Central Europe. The same species were found for example in Great Britain (Khan, 1960) or Finland (Niewiadomska *et al.*, 1997); this occurrence is connected with the large distribution area of the great pond snail in Europe.

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