

Four New Xiphidiocercariae from *Melanopsis praemorsa* (L., 1758) (Thiaridae) Snails in Jordan

NAIM S. ISMAIL AND SAMI M. BDAIR

(Received for publication; April 4, 1986)

Abstract

Four new types of xiphidiocercariae are described from the freshwater snail *Melanopsis praemorsa* collected from several freshwater bodies in Jordan: Two microcotylous non-virgulate xiphidiocercariae belonging to the "Pusilla" sub-type, and two virgulate xiphidiocercariae. These cercariae are named *Cercaria melanopsi* X through XIII. Details are presented on the morphology and the behaviour of the cercariae as well as on their development within the snail.

Key words: Xiphidiocercariae, cercariae, larval trematodes, *Melanopsis*, Jordan

Introduction

Melanopsis praemorsa snail is prevalent in the Mediterranean region (Tchernov, 1975; Brown and Wright, 1980). In Jordan it is common in springs, streams, and rivers (Schutt, 1983). Several types of cercariae have been described from *M. praemorsa* snails. Of these, six types were xiphidiocercariae: *Cercaria melanopsi* I, II and III (Ismail and Abdel-Hafez, 1983), *C. melanopsi* IX (Ismail and Abdel-Hafez, 1984), and *Cercaria Levantina* 3 and 4 (Lengy and Stark, 1971). This paper describes additional new four types of xiphidiocercariae from *M. praemorsa* in Jordan.

Materials and Methods

A total of 15,918 *M. praemorsa* snails were collected from 32 freshwater bodies in Jordan during the period from April 1984 to October 1985. Collected snails were isolated individually in Syracuse dishes and the water was examined the following morning. Both snails that had shed cercariae and those that had not were crushed, dissected, and examined for developing larval stages. Observations on recovered larval stages were made on live, unstained or

vitaly stained (0.5% neutral red or 0.5% brilliant cresyl blue) specimens, as well as on specimens fixed in FAA (foramlin-acetic acid-alcohol) solution and subsequently stained in acetocarmine.

Larval morphometrics were taken from at least 10 live and 10 fixed specimens. Camera lucida drawings from fixed specimens were completed free hand from observations on live specimens using a compound microscope. The behaviour of cercariae was observed using a dissecting microscope.

Results

Four new types of xiphidiocercariae were found in *M. praemorsa*: two virgulate and two non-virgulate xiphidiocercariae. They are named *Cercaria melanopsi* X, *C. melanopsi* XI, *C. melanopsi* XII, and *C. melanopsi* XIII. The morphometrics of these cercariae are shown in Table 1.

Cercaria melanopsi X: (Figs. 1A, B, C)

This cercaria was found in 11 out of 1371 snails collected from Al-Hammeh and Al-Hawi sites at Yarmouk River and from Wadi Ziglab. It is relatively small cercaria and is covered with a 1 μ m tegument which is provided with fine spines. Six long sensory setae (about 3 μ m long) are also present on each side of the body. The

Table 1 Morphometric measurements, in microns, of the *Cercaria melanopsi* X through XIII

Morphometric Measurement	Type of <i>Cercaria melanopsi</i>			
	X	XI	XII	XIII
Body length*	68—83 60—80	93—140 100—135	105—158 118—135	105—150 115—140
Body width*	35—58 30—40	58—93 55—80	58—85 68—80	38—80 55—73
Tail length*	65—80 33—90	73—90 38—88	38—138 38—68	63—83 53—83
Tail width*	10—11 10—13	10—15 15—25	13—15 20—25	10—13 15—20
Oral sucker†	25 × 22	28 × 28	38 × 63	50 × 35
Acetabulum†	18 × 15	17 × 16	20 × 20	18 × 17
Pharynx†	8 × 7	7 × 7	10 × 10	5 × 5
Stylet† (length × width)	14 × 4	16 × 4	25 × 3	18 × 2.3

*Measurements are given for live (upper values) and fixed (lower values) specimens.

†Measurements are given for live specimens.

oral sucker is oval in shape and is relatively small. It is provided with a stylet, but lacks the virgula organ. The spherical acetabulum is smaller than the oral sucker and is located in the posterior half of the body. The digestive system consists of a mouth that leads posteriorly into an oval pharynx, which in turn connects to a short esophagus. Intestinal caeca were not detected. The excretory system consists of a rectangular excretory bladder measuring 5 μm long by 10 μm wide and tends to be bicornuate when it is fully distended. The excretory bladder receives two primary excretory canals. Secondary and tertiary excretory canals were not detected. The bladder is connected to a caudal excretory canal that opens into a terminal excretory pore at the posterior end of the tail. A total of 26 flame cells are found distributed symmetrically on both sides of the body, but are absent in the tail. Genital primordium consists of a mass of cells immediately behind the acetabulum. Round cystogenous cells (about 2 μm in diameter) are scattered throughout the body. The glandular system is made up of three pairs of penetration glands on both sides of the body. They are of the same size, have coarse granules, and are arranged symmetrically around the acetabulum. Two

pairs of glands pour their contents into ducts which run anteriorly to open on both sides of the stylet. The third pair of glands have ducts that open at the inner margin of the oral sucker.

This cercaria is an active swimmer. It swims in all directions by the movement of the tail. It does not reach the water surface. Once it is near the surface it pauses and starts sinking slowly to the bottom. When on the bottom, the cercaria stands still for a while before it suddenly starts to swim again. Cercariae show positive thigmotaxis, tending to concentrate along the edge of the dish. The emerged cercariae usually die within 24 hours without encystment.

This cercaria develops within an oval shaped sporocyst. Sporocysts measure 210—370 by 120—210 μm . One end of the sporocyst is usually plug-shaped. It is most probably the birth pore. Sporocysts usually contain up to five developed cercariae, 4—20 developing cercariae, and germ balls.

Cercaria melanopsi XI: (Fig. 1 D, E, F, G)

This cercaria was found in 10 out of 278 snails collected from Ghazal spring. Its body is covered with 1—3 μm thick tegument which is covered with fine spines. Longer spines are con-

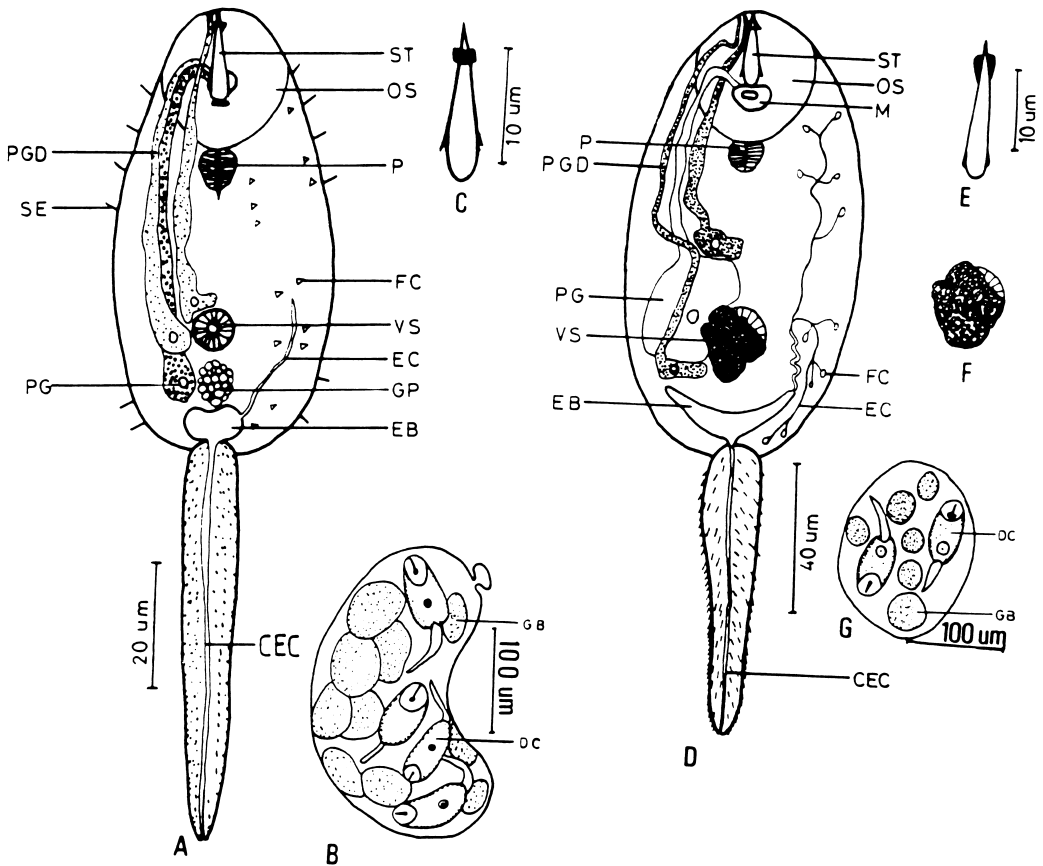


Fig. 1 Non-irgulate xiphidiocercariae encountered in *Melanopsis praemorsa* snails from Jordan. A–C, *Cerariae melanopsi* X and its stylet and sporocyst. D–G, *Cerariae melanopsi* XI and its stylet, sporocyst, and ventral sucker covered with the genital primordium.

Abbreviations

CED: Caudal excretory canal, DC: Developed cercaria, EB: Excretory bladder, EC: Excretory canal, ES: Esophagus, FC: Flame cell, GB: Germ ball, GP: Genital primordium, M: Mouth, OS: Oral sucker, P: Pharynx, PG: Penetration gland, PGD: Penetration gland duct, SE: Seta, ST: Stylet, VO: Virgula organ, VS: Ventral sucker

centrated on the tail. The oral sucker is spherical in shape and has a stylet but lacks a virgula organ. The spherical acetabulum is smaller than the oral sucker and is located in the posterior half of the body at about $70\ \mu\text{m}$ from the anterior extremity. The digestive system consists of a mouth that leads posteriorly into a pharynx abutting the posterior border of the oral sucker. No intestinal caecum was detected. The excretory system consists of a bicornuate excretory bladder located centrally at the posterior end of the body. It opens into a terminal

excretory pore at the posterior tip of the tail. The bladder receives two primary excretory ducts which are convoluted near the bladder. Each duct bifurcates into two secondary canals at the level of the acetabulum, one runs anteriorly to about the level of the oral sucker and the other runs posteriorly to the end of the body. Flame cells are arranged symmetrically in the body and are absent in the tail. The flame cell formula is $2[(2+2+1) + (2+2+2)] = 22$. The genital primordium consists of a mass of cells covering the acetabulum and the area between

the acetabulum and excretory bladder. The glandular system is made up of three pairs of penetration glands on both sides of the body and around the acetabulum. The upper gland is relatively small with coarse granules. The lower gland is the smallest with granules finer than those of the upper gland. The largest gland is located in between the two mentioned glands and it is agranulated. Glands with coarse granules pour their contents into ducts that open at both sides of the stylet. The large gland has a duct that opens at the inner edge of the oral sucker.

This cercaria is an active swimmer. It swims in all directions by the action of the tail, but does not reach to the surface. When it stops swimming it sinks slowly to the bottom on which it rests. This cercaria does not crawl on the bottom. Cercariae show positive thigmotaxis and positive phototaxis. The emerged cercariae usually die within 24 hours without

encystment.

This cercaria develops within spherical shaped sporocysts. They measure 163–238 by 150–175 μm . Sporocysts usually contain 1–3 developed cercariae, 1–6 developing cercariae, and germ balls.

Cercaria melanopsi XII: (Fig. 2 D, E, F)

This cercaria was found in 90 out of 1,982 snails collected from Al'al, Tais, Karam, Foutaha, and Sayegh springs, and from Wadi Ziglab. Its body is covered with 2–3 μm thick tegument which bears fine spines. Eight long setae are also distributed on each side of the body. The tail is highly contractile and covered with fine spines. The oral sucker is relatively large. It bears two characteristic structures, the stylet which is dagger-shaped and protrudes outside the edge of the body, and the virgula organ which is bilobed and covers about two-thirds of the oral sucker. The virgula organ contains non-

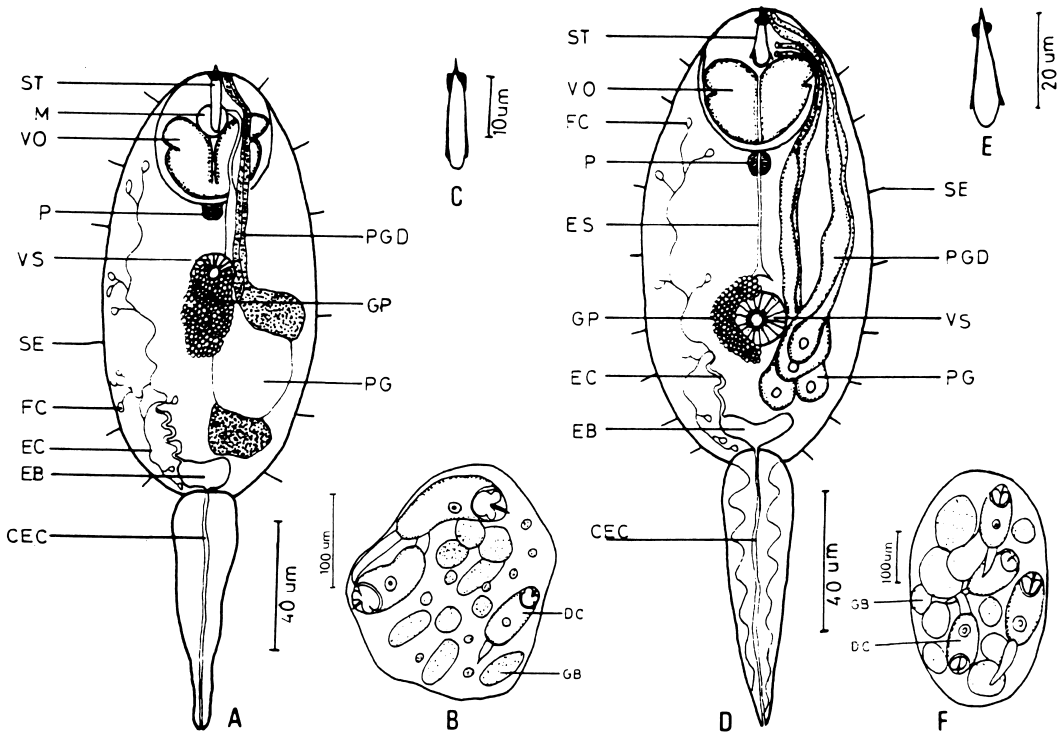


Fig. 2 Virgulate xiphidiocercariae encountered in *Melanopsis praemorsa* snails from Jordan. D–F, *Cercaria melanopsi* XII and its stylet and sporocyst. A–C, *Cercaria melanopsi* XIII and its stylet and sporocyst.

granulated material. The ellipsoidal acetabulum is smaller than the oral sucker and is located in the posterior half of the body at about $67\ \mu\text{m}$ from the anterior extremity. The digestive system consists of a mouth that leads posteriorly into a pharynx abutting the posterior border of the oral sucker. The pharynx opens into a long esophagus that bifurcates near the acetabulum into two short intestinal caeca. The excretory bladder is relatively large measuring 25 by $20\ \mu\text{m}$. It tends to be bicornuate when it is fully distended. It is located centrally at the posterior end of the body and opens into a terminal excretory pore at the posterior end of the tail. The bladder receives two primary excretory ducts which are convoluted near the excretory bladder. Each duct bifurcates into two secondary excretory ducts shortly behind the acetabulum. One duct runs anteriorly to about the level of the oral sucker and the other runs posteriorly to the posterior end of the body. Flame cells are arranged symmetrically on both sides of the body. The flame cell formula is $2[(3+2) + (1+3)] = 18$. The genital primordium consists of a mass of cells converging one half of the acetabulum. The glandular system is made up of four pairs of penetration glands on both sides of the body. Glands fill the space between the acetabulum and the excretory bladder. Glands pour their granular contents into four pairs of ducts that run anteriorly. Two pairs open on both sides of the stylet and two pairs open at the inner edge of the oral sucker. Cystogenous cells are concentrated on the periphery of the body. The cercaria undergoes a partial encystment under the pressure of the cover slip.

This cercaria is an active swimmer. It swims near the bottom of the dish, sometimes it rests on the bottom, then starts crawling on it by the action of body contractions. In this case the cercaria uses its acetabulum for positioning. The cercaria can swim upward into the water column and when stops swimming starts to sink slowly. Thus, many cercariae were seen hanging in the water. The body of the cercaria contracts also while the cercaria is hanging in the water. Cercariae usually die within 24 hours without encystment.

This cercaria develops within a spherical shaped sporocyst. Sporocysts measure $350\text{--}530$ by $150\text{--}300\ \mu\text{m}$. They usually contain 1–4 developed cercariae, 1–4 developing cercaria, and germ balls.

Cercaria melanopsi XIII: (Fig. 2 A, B, C)

This cercaria was found in 8 out of 695 snails collected from Beida, Abu-Ziyad, Zughdiya Springs, and from Wadi Ziglab. Its body is covered with a $1\text{--}2\ \mu\text{m}$ thick tegument which is covered with fine spines. A total of six sensory setae are distributed on each side of the body. The oral sucker bears two characteristic structures, the stylet which protrudes outside the edge of the body, and the virgula organ which is bilobed and covers about two-thirds of the oral sucker. The virgula organ contains non-granulated material. The acetabulum is located at the middle of the body and usually protrudes laterally. The digestive system consists of a mouth that leads posteriorly into a muscular pharynx abutting the posterior border of the oral sucker. No intestinal caecum was detected. The excretory system is conventional. The excretory bladder is round and is located centrally at the posterior end of the body. It opens into a terminal excretory pore at the tip end of the tail. The bladder receives two primary excretory ducts which are convoluted near the excretory bladder. Each duct bifurcates into two secondary ducts, one that runs anteriorly to about the level of the oral sucker, and the other runs posteriorly to the end of the body. Flame cells are arranged symmetrically on both sides of the body. The flame cell formula is $2[2+2+1] + (3+1+2) = 22$. The genital primordium consists of a dense cluster of cells located around the acetabulum and between the acetabulum and the excretory bladder. The glandular system is made up of three pairs of penetration glands on both sides of the body between the acetabulum and the excretory bladder. The upper gland has coarse granules, the lower has fine granules, and the middle is the largest and is agranulated. The contents of the upper and lower glands is transported through two pairs of ducts that open on both sides of the stylet. The

agranulated gland has a duct that runs anteriorly and opens at the inner margin of the oral sucker.

This cercaria is an active swimmer. It swims near the water surface. When it stops swimming it sinks slowly and can re-swim before reaching the bottom. Although the body of the cercaria contracts on the bottom, the cercaria does not crawl actively on the bottom. Cercariae show positive thigmotaxis and positive phototaxis. The emerged cercariae usually die within 24 hours without encystment.

This cercaria develops within spherical sporocysts measuring 170–320 by 120–270 μm . Sporocysts usually contain 1–4 developed cercaria, 1–5 developing cercariae, and germ balls.

Discussion

The presence of well developed stylets in *Cercaria melanopsi* X, XI, XII and XIII identifies them as xiphidiocercariae. In *C. melanopsi* XI, the absence of the tail finfold, the small size of the body, and the location and the size of the acetabulum relate them to the microcotylae subgroup of the xiphidiocercariae as described by Lühe (1909). The possession of the three pairs of penetration glands, undeveloped gut, lack of virgula organ in the oral sucker, and the development within small ovoid sporocysts indicate that these cercariae belong to the "Pusilla" sub-type as described by Sewell (1922). The other species of non-virgulate xiphidiocercariae which resemble *C. melanopsi* X and XI and possess three pairs of penetration glands, lack a caudal finfold and eyespots, and lack intestinal caeca are: *C. indicae* XVIII, XIV, XL and XLVI (Sewell, 1922), *Cercaria Levantina* 13 (Gold and Lengy, 1974); *C. melanopsi* I (Ismail and Abdel-Hafez, 1983). *Cercaria melanopsi* X can be differentiated from all of these cercariae on the basis of its small size, the position and arrangement of penetration glands, the shape and size of the stylet, and the number of flame cells. Although *C. melanopsi* X closely resembles *C. melanopsi*

I, they are different in several aspects: the stylet of *C. melanopsi* X has more enforcement than *C. melanopsi* I, and the penetration glands of *C. melanopsi* X reach down to about the level of the excretory bladder while they do not in *C. melanopsi* I. In addition *C. melanopsi* X has 26 flame cells while *C. melanopsi* I has 20. Similarly *C. melanopsi* XI can be differentiated from all of the above mentioned cercariae on the basis of the position and the arrangement of penetration glands, the shape and size of stylet, and the number of flame cells. Moreover, the genital primordium of *C. melanopsi* XI is characteristically covering most of the ventral sucker.

In *C. melanopsi* XII and XIII, the presence of virgula organs indicate that they belong to Cercariae Virgulae as described by Lühe (1909). Cercariae Virgulae accommodate the forms with a characteristic virgula organ, a ventral sucker smaller than the oral, three to six pairs of penetration glands, excretory bladder V-shaped, oval or reniform, and tail without a finfold. Sewell (1922) established two subgroups of Cercariae Virgulae: Virgula subgroup for cercariae of small size (body less than 0.2 mm), and lacking esophagus and caecum; and Paravirgula subgroup for cercariae with esophagus and caeca. The presence of intestinal caeca behind the pharynx in *C. melanopsi* XII indicates that it belongs to "Paravirgula" group described by Lühe (1901), while the absence of such caeca in *C. melanopsi* XIII indicates that it belongs to the "Virgula" group.

The other species of virgulate xiphidiocercariae which resemble *C. melanopsi* XII and possess four pairs of penetration glands and an intestinal caeca are *Cercaria stenophenezua* (Nasir and Diaz, 1973), *C. microvirgula* (Haseeb and Khan, 1984), and *Cercaria tarda* (Khan, 1961). *Cercaria melanopsi* XII can be differentiated from all these cercariae on the basis of the size and shape of the virgula organ, the shape of the stylet, and the number of flame cells. *Cercaria melanopsi* XII closely resembles *C. tarda*. However, the latter has 24 flame cells versus 18 in the former. Although *C. tarda* has

a bilobed virgula organ as *C. melanopsi* XII, the shape of the virgula organ of both is different. This is in addition to the fact that *C. melanopsi* XII is smaller than *C. tarda*.

The other species of virgulate xiphidocercariae which resembles *C. melanopsi* XIII and possess three pairs of penetration glands, lack a caudal fin-fold and eyespots, and lack intestinal caeca are: *C. melanopsi* II, III, IX (Ismail and Abdel-Hafez, 1983, 1984), *C. indicae* XXXIV, XLIV, XLV and LVI (Sewell, 1922). *Cercaria melanopsi* XIII can be differentiated from all these cercariae on the basis of the shape and size of the stylet, the size and arrangement of the penetration glands, and the shape of the virgula organ. Although *C. melanopsi* XIII closely resembles *C. melanopsi* IX, the penetration glands of the latter is smaller and are arranged around and slightly behind the acetabulum, while the former has penetration glands that extend down to about the excretory bladder. Moreover, *C. melanopsi* IX has 18 flame cells, while *C. melanopsi* XIII has 22.

Acknowledgments

This work was supported by a grant from Yarmouk University Research Council. The authors wish to thank Dr. Sami K. Abdel-Hafez for his valuable comments and suggestions. The help of Mr. Issam Issa in the field and laboratory is appreciated.

References

- 1) Brown, D. S. and Wright, C. A. (1980): Mollusca of Saudi Arabia: freshwater molluscs. Fauna of Saudi Arabia, 2, 341–358.
- 2) Gold, D. and Lengy, J. (1974): Studeis on larval stages of digenetic trematodes in aquatic molluscs of Israel. 4. On five cercariae from the freshwater snail *Melanoides tuberculata* (Müller, 1774). Isr. J. Zool., 23, 143–161.
- 3) Haseeb, M. A. and Khan, D. (1984): Studies on larval trematodes infecting freshwater snails in Pakistan. IX. Virgulate xiphidocercariae. Proc. Helminthol. Soc. Washington, 51(2), 282–286.
- 4) Ismail, N. S. and Abdel-Hafez, S. K. (1983): Larval stages of digenetic trematodes of *Melanopsis praemorsa* (L. 1758, Buccinum) (Thiaridae) snails from Yarmouk River, Jordan. Z. Parasitenkd., 69, 613–626.
- 5) Ismail, N. S. and Abdel-Hafez, S. K. (1984): Two new cercariae from *Melanopsis praemorsa* (L., 1758) (Thiaridae) snails in Azraq Oasis, Jordan. Jpn. J. Parasitol., 33(4), 353–359.
- 6) Khan, D. (1961): Studies on larval trematodes infecting freshwater snails in London (U.K.) and some adjoining areas. Xiphidocercariae. Z. Parasitenkd., 21, 71–87.
- 7) Lengy, J. and Stark, A. (1971): Studies on larval stages of digenetic trematodes in aquatic molluscs of Israel. 2. On three cercariae encountered in the freshwater snail *Melanopsis praemorsa* L. Isr. J. Zool., 20, 41–51.
- 8) Lühe, M. (1909): Parasitische Plattwürmer. I. Trematoden. Süßwass. Fauna Dtl., 17, 1–215.
- 9) Nasir, P. and Diaz, M. T. (1973): Freshwater larval trematodes. XXXII. Twenty new species of Venezuelan cercariae. Riv. Parasitologia, 34(1), 1–44.
- 10) Schutt, H. (1983): Die bisher aus Jordanien bekannten süßwasser- und landbewohnende Mollusken anhand der Aufsammlungen von. Dr. B. Nandel, 1978. Natur. und Mensch., 49–64.
- 11) Sewell, R.B.S. (1922): Cercariae Indicae. Indian J. Med. Res. 10 (Suppl.), 1–370.
- 12) Tchernov, E. (1975): The mollusca of the Sea of Galilee. Malacologia, 15, 147–184.