

**STUDIES ON THE LARVAL TREMATODES
(DIGENEA) WITH EMPHASIS ON CERCARIAE
INFECTING FRESHWATER SNAILS IN
KERALA, INDIA**

*Thesis submitted to the
University of Calicut for the Degree of*

**DOCTOR OF PHILOSOPHY
IN ZOOLOGY**

By

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CERTIFICATE

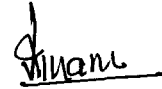
This is to certify that this thesis is an authentic record of the research work done by **Sri. Vasandakumar, M.V.** from August 1995 to November 2002 under my supervision and guidance.

Dr. K.P. Janardanan

DECLARATION

I hereby declare that the thesis entitled **Studies on the larval trematodes (Digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India** has been carried out by me under the guidance of Prof. (Dr.) K.P. Janardanan, Department of Zoology, Calicut University and that this work has not been published or submitted in part or full for any degree or prize.

Calicut University,
19.11.2002.



Vasandakumar, M.V.

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INTRODUCTION

Vasandakumar M.V. “Studies on the larval trematodes (digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India” Thesis. Department of Zoology , University of Calicut, 2002

INTRODUCTION

Digenetic trematodes form an important group of helminth parasites because they parasitize most of the vertebrates, and many of them cause serious diseases to man and animals of economic importance. In India, extensive work has been carried out on the trematode fauna of various vertebrates. Compared to the bulk of knowledge available on adult trematodes, only very little information is available on their larval stages and life cycles. Information on cercariae serves as one of the most dependable bases for the taxonomy of trematodes, and specific identity of cercariae with adults can be made by raising the adults from cercariae. Sewell in 1922 took up the study of larval trematodes of India and published a monograph on Indian cercariae. Since then no serious efforts have been made to explore further the cercarial fauna of the entire country. However, a few workers have paid attention to explore the cercarial fauna of a particular zone or state. My preliminary studies revealed that probably few, if any, species of snails in the Malabar area (comprising Kasaragod, Kannur, Wayanad, Kozhikode, Malappuram and Palakkad districts) are totally free from larval trematode infections. Since relatively little is known about the cercarial fauna of this region, the present investigation has been undertaken with a view to contributing to the knowledge of the cercarial fauna of Malabar, Kerala.

During the present study, freshwater snails belonging to 8 genera and 10 species were examined, and as many as 22 species of cercariae, including 11 new species, were recovered from 6 species of snails. Complete life cycles have been traced for two species of digeneans, *Plagioporus panchax* n. sp. and *Petasiger variospinosus*. Partial life cycle of a species of *Microparaphium* has been worked out using cercaria as the starting point. Cercariae of known life cycles are referred to by their adult names. The new species of cercariae are respectively designated as *Cercaria* sp. I (to XI) Malabar series to indicate the region of the collection of snails. Detailed descriptions are presented for all the new species. La Rue (1957) has been followed in the arrangement of cercariae. Data on prevalence of infections by various cercariae have been collected and presented. Host specificity and other aspects of host-parasite relations of cercariae have been discussed in the light of available information.

HISTORICAL REVIEW

Vasandakumar M.V. “Studies on the larval trematodes (digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India” Thesis. Department of Zoology , University of Calicut, 2002

HISTORICAL REVIEW

The early 18th century marks the beginning of the study of larval trematodes. In 1773, it was O.F. Muller who for the first time introduced the term 'Cercaria' to describe the microscopic animals with tails. The term was used as a generic name since the larval stages were considered to be adult trematodes and no one suspected about the need for a change of hosts for the completion of their life cycles. Subsequently, Abildgaard (1790) described a larval trematode considering it to be an adult organism. In the early 19th century also cercariae were described as independent organisms of unknown taxonomic position. It was only after Steenstrup's studies in 1842 that cercariae were regarded as larvae of organisms parasitic in vertebrate animals. He showed that cercariae are trematode larvae and attain sexual maturity only after their entry into the intestine of vertebrate animals. Further, he assumed that cercariae arise in sporocysts (or rediae) as a result of budding and the life cycle of trematodes involves an alternation of sexual and asexual generations. Diesing (1850) still considered cercariae to be adult trematodes, and erected a suborder for them. His revision of the cercariae in 1855 recognized 20 species and 9 genera. Later, he (1858) came to accept the larval nature of cercariae but continued to use generic names for them.

Since then, many workers began to focus their attention on the study of the life history of digeneans by observations and ill-devised experiments, and these met

with varying degrees of success. In 1854 Siebold demonstrated experimentally the connection between a number of intestinal worms and the larval stages. Leuckart (1882) and Thomas (1883) for the first time worked out independently the life cycle of the sheep liver fluke, *Fasciola hepatica* from Germany and Great Britain respectively. This became a land mark in the history of life cycle studies of digenetic trematodes and paved the way for subsequent studies on cercarial fauna of snails and life cycles of digenetic trematodes. In 1889 and 1892, Braun reviewed the literature dealing with cercaria and life histories of digeneans accumulated during the period 1843-1892, and listed more than 63 titles of research papers. Without doubt, the most significant studies in this period were those made by Leuckart (1882) and Thomas (1883). Looss (1892) attempted to trace the relationship between the amphistome cercaria, *Cercaria diplocotylea* and the frog trematode *Diplodiscus subclavatus*. Sonsino in 1892 described another amphistome cercaria *Cercaria pigmentata* and Looss (1896) showed that it developed into *Amphistomum conicum* (= *Paramphistomum cervi*). Sonsino in the same year described the first freshwater larval trematode from Egypt. Through the studies of Osafune (1897-1898), fragmentary information on Japanese cercariae began to appear, and Seno in 1903 reported several species of cercariae in Japan.

With the increase in knowledge, the importance of larval characters for taxonomy of digenetic trematodes has been realised by many workers. Luhe in 1909 made the first serious attempt to classify cercariae based on external

characters such as suckers, stylets, collar and collar spines and tails. Lebour (1911) made an alternative attempt to classify cercariae emphasising the mode of origin of cercariae. La Rue (1957) proposed a system of classification mostly based on larval characters particularly the excretory system of cercariae.

The twentieth century witnessed enormous number of publications dealing with the description of many species of cercariae and the discovery of large number of life cycles. It is very difficult to cite here all the works but the relevant ones are referred to elsewhere at the appropriate places. Mention must, however, be made here of some of the valuable contributions made by Cort (1914, 1915), Faust (1918, 1919, 1924), Harper (1929), Krull (1934), Bennet (1936), Porter (1938), Bissuru (1953, 1967), Nasir (1960, 1964, 1980), Ching (1961), Lie (1963, 1964, 1965), Lie and Umathevy (1965, 1966), Lie and Basch (1967), Hsu *et al.* (1968), Lengy and Wolff (1971), Ito (1977a,b,c), Blair (1977), Ito *et al.* (1977a,b), Ito and Blas (1978), Lengy and Gold (1978), Khan and Haseeb (1981), Kanev and Odening (1983), Haseeb (1984), Ostrowski de Nunez (1984, 1986, 1987), Whyte *et al.* (1988), Ismail (1990), Ismail and Arif (1991), Abdulsalam and Sreelatha (1993, 1994, 1996), Kanev (1994), Kanev *et al.* (1995), Ostrowski de Nunez *et al.* (1996), Fried *et al.* (1998), Haberl *et al.* (2000), Abrous *et al.* (2000) and others on the cercariae and/or life cycles of Digenea from abroad. Yamaguti (1975), in a synoptical review summarised much of the information on the life cycles of digenetic trematodes.

In India, larval trematodes infecting molluscs in various water bodies have been a subject of serious research for more than a century. Information is also available on life cycles of these larval stages although there are still many gaps. Liston and Soparkar (1918) were the pioneers in this field; they began with the life history of *Schistosoma spindalis*. Later, Soparkar (1921a,b) published a full account of the cercaria of *S. spindalis* and also a few other furcocercous cercaria in *Planorbis exustus* in Bombay. Sewell conducted an extensive study on the cercarial fauna of India and published his monumental work, 'Cercariae Indicae' in the year 1922. In this monograph, he described 62 species of cercariae occurring in several parts of India. This is the most detailed work on the cercarial fauna of India. After this classical work, several publications have appeared on freshwater cercariae, some dealing with reports of individual cercariae and others devoted to more comprehensive surveys. Important among them are the contributions made by Rao (1932a), Peter and Mudaliar (1948), Anantaraman and Balasubramaniam (1949), Singh (1952a, 1953, 1955a), Premvati (1953a, 1955, 1956), Peter (1955a, 1956), Murty (1966, 1975a,b, 1976), Pandey (1967a,b, 1973a,b,c), Srivastava (1968), Ganapati and Rao (1969, 1983), Gupta and Taneja (1969a,b), Thapar (1969), Pandey and Srivastava (1970), Agrawal (1974), Baugh (1975), Mukherjee and Ghosh (1977), Saxena (1977), Jain and Sharma (1981), Agrawal and Sharma (1980), Ramachandrupa and Agarwal (1985, 1987), Singh and Bali (1991), Srivastava and Saxena (1992a,b), Agarwal *et al.* (1993a,b), Dhanumkumari *et al.*

(1993), Meenakshi *et al.* (1993), Hyalij and Deoray (1995), Umadevi and Madhavi (1995), Choubisa (1997, 1998) and Pandey and Panwar (2000). Under the "Fauna of India" series Mukherjee (1986) gave an up to date account on amphistome cercariae, and in 1992 on parapleurolophocerca and echinostome cercariae.

Information on life cycles of Indian larval trematodes has also accrued slowly. The notable contributions were made by Singh (1959a,b), Ramalingam (1960), Patnaik and Ray (1966), Rao and Ganapati (1967), Murty (1973), Madhavi (1978a,b, 1980b), Madhavi and Shameem (1986), Madhavi *et al.* (1987), Shameem and Madhavi (1988), Madhavi and Swarnakumari (1995), Madhavi and Rukmini (1997), Jhansilakshmibai and Madhavi (1997) and Umadevi and Madhavi (2000). There are several other workers also who have contributed much to this field of study.

Digenetic trematodes in Kerala also have attracted the attention of a few research workers. Sewell (1922), in his monograph, 'Cercariae Indicae', reported the occurrence of 32 species of cercariae from 10 species of freshwater snails of Kerala. Later, Nadakal *et al.* (1969), Mohandas (1971a, 1973a,b, 1974a,b, 1976, 1977, 1981), Janardanan *et al.* (1987), Janardanan and Shiny (1989), Janardanan and Prasadani (1991), Rajendran and Janardanan (1993), Roopa and Janardanan (1998, 2001) and Nambiar and Janardanan (2001) reported several cercariae and elucidated the life cycles of 5 species of trematodes in Kerala.

Several species of cercariae infecting freshwater snails have been reported from different parts of India. As it is very difficult to list out all of them here, I am presenting separate lists of cercariae recorded so far from the snails, *Bellamya* spp. (Table 1), *Pila* spp. (Table 2), *Digoniostoma* spp. (Table 3), *Thiara* spp. (Table 4), *Paludomus* spp. (Table 5), *Lymnaea* spp. (Table 6), *Indoplanorbis* spp. (Table 7) and *Gyraulus* spp. (Table 8), which were examined during the course of the present investigation.

Table 1. List of cercariae reported from *Bellamyia* spp. in India

Sl. No.	Name	Locality	Reference
1	2	3	4
1.	<i>Cercariae indicae</i> LXII Sewell, 1922	Calcutta	Sewell (1922)
2.	<i>Cercaria pendulata</i> Baugh, 1954	Lucknow	Baugh (1954a)
3.	<i>C. lucknowensis</i> Baugh, 1954	Lucknow	Baugh (1954b)
4.	<i>C. kumaunensis</i> Singh and Malaki, 1963	Kumaun, Kakinada	Singh and Malaki (1963), Murty (1975a)
5.	<i>C. esplanadensis</i> Mukherjee and Ghosh, 1975	Esplanade, Calcutta	Mukherjee and Ghosh (1975, 1982)
6.	<i>C. beaverii</i> Pandey and Agarwal, 1977	Lucknow	Pandey and Agarwal (1977)
7.	<i>C. komiyai</i> Pandey and Agarwal, 1977	Lucknow	Pandey and Agarwal (1977)
8.	<i>C. chauhani</i> Pandey, 1975	Lucknow	Pandey (1975)
9.	Cercaria of <i>Azygia papillata</i> Ubgade and Agarwal, 1980	Chikhali, Raipur	Ubgade and Agarwal (1980)
10.	<i>Cercaria baniensis</i> Jain, Pandey and Sharma, 1982	Lucknow	Jain <i>et al.</i> (1982)
11.	<i>C. udaipuriensis</i> Sharma and Choubisa, 1983	Sagar Lake	Sharma and Choubisa (1983)
12.	<i>C. spinosa</i> Pandey and Singh, 1984	Unnao	Pandey and Singh (1984)

1	2	3	4
13.	<i>C. chauhani</i> Pandey and Singh, 1986	Lucknow	Pandey and Singh (1986)
14.	<i>C. premvatai</i> Pandey and Singh, 1986	Lucknow	Pandey and Singh (1986)
15.	<i>C. srivastavi</i> Pandey and Singh, 1986	Lucknow	Pandey and Singh (1986)
16.	<i>C. chiraiyabaghensis</i> Saxena and Srivastava, 1992	Lucknow	Saxena and Srivastava (1992)
17.	<i>C. bellamayii</i> Srivastava and Saxena, 1992	Lucknow	Srivastava and Saxena (1992)
18.	<i>C. laungakheraii</i> Srivastava and Saxena, 1992	Lucknow	Srivastava and Saxena (1992)

Table 2. List of cercariae reported from *Pila* spp. in India

Sl. No.	Name	Locality	Reference
1.	<i>Cercaria andraensis</i> Ganapati and Rao, 1969	Waltair	Ganapati and Rao (1969)
2.	Cercaria of <i>Pseudodiplodiscoides pilai</i> Murty, 1973	Kolleura lake	Murty (1973)
3.	<i>Cercaria pilai</i> I Janardanan and Shiny, 1989	Idimuzhikkal	Janardanan and Shiny (1989)
4.	<i>C. pilai</i> II Janardanan and Shiny, 1989	Idimuzhikkal, Thenhippalam	Janardanan and Shiny (1989)
5.	Cercaria of <i>Tremiorchis ranarum</i> Mehra and Negi, 1926	Chelembra	Rajendran and Janardanan (1993)

Table 3. List of cercariae reported from *Digoniostoma* spp. in India

Sl. No.	Name	Locality	Reference
1	2	3	4
1.	<i>Cercariae indicae</i> V Sewell, 1922	Calcutta	Sewell (1922)
2.	<i>C. indicae</i> VIII Sewell, 1922	Calcutta	Sewell (1922)
3.	<i>C. indicae</i> XXXI Sewell, 1922	Calcutta	Sewell (1922)
4.	<i>C. indicae</i> XXXII Sewell, 1922 (= Cercaria of <i>Ceylonocotyle scoliocoelum</i> (Fischoeder, 1904) Näsmark, 1937)	Calcutta, Bareilly	Sewell (1922), Mukherjee (1960a), Jain <i>et al.</i> (1971)
5.	<i>C. indicae</i> LVII Sewell, 1922	Calcutta	Sewell (1922)
6.	<i>C. indicae</i> LIX Sewell, 1922	Calcutta	Sewell (1922)
7.	<i>C. indicae</i> LX, Sewell, 1972	Calcutta	Sewell (1922)
8.	<i>C. indicae</i> LXI Sewell, 1922	Calcutta	Sewell (1922)
9.	<i>Cercaria pulchella</i> Mukherjee, 1963	Bareilly	Mukherjee (1963)
10.	<i>C. kareilliensis</i> Mukherjee, 1972	Bareilly	Mukherjee (1972)
11.	<i>Cercariae indicae</i> LXXI Murty, 1975	Waltair	Murty (1975c)
12.	<i>Cercaria</i> sp. IV Kerala Mohandas, 1976	Trivandrum	Mohandas (1976)
13.	<i>Cercaria</i> sp. V. Kerala Mohandas, 1977	Trivandrum	Mohandas (1977)

1	2	3	4
14.	<i>Cercaria</i> sp. XII Kerala Mohandas, 1977	Trivandrum	Mohandas (1977)
15.	<i>C. raipurensis</i> VI Ramachandrupala and Agarwal, 1985	Raipur	Ramachandrupala and Agarwal (1985)
16.	<i>C. raipurensis</i> V Ramachandrupala and Agarwal, 1987	Raipur	Ramachandrupala and Agarwal (1987)
17.	<i>C. raipurensis</i> XII Ramachandrupala and Agarwal, 1990	Lucknow	Ramachandrupala and Agarwal (1990)
18.	<i>Cercaria</i> of <i>Pleurogenoides</i> <i>ovatus</i> Rao, 1977	Malappuram	Janardanan and Prasadana (1991)

Table 4. List of cercariae reported from *Thiara* spp. in India

Sl. No.	Name	Locality	Reference
1	2	3	4
1.	<i>Cercariae indicae</i> III Sewell, 1922	Calcutta, Bombay	Sewell (1922)
2.	<i>C. indicae</i> IV Sewell, 1922	Wayanad, Calcutta, Bombay	Sewell (1922)
3.	<i>C. indicae</i> V Sewell, 1922	Calcutta	Sewell (1922)
4.	<i>C. indicae</i> VI Sewell, 1922	Calcutta	Sewell (1922)
5.	<i>C. indicae</i> VII Sewell, 1922	Calcutta, Bombay	Sewell (1922)
6.	<i>C. indicae</i> VIII Sewell, 1922 (= <i>Cercaria</i> of <i>Acanthostomum burminis</i> (Bhalerao, 1926) Bhalerao, 1936)	Calcutta, Bombay, Nilambur, Kannur, Kozhikode, Malappuram	Sewell (1922), Mohandas (1976), Roopa and Janardanan (1998)
7.	<i>C. indicae</i> XI Sewell, 1922	Calcutta	Sewell (1922)
8.	<i>C. indicae</i> XIII Sewell, 1922	Calcutta	Sewell (1922)
9.	<i>C. indicae</i> XIV Sewell, 1922	Calcutta	Sewell (1922)
10.	<i>C. indicae</i> XV Sewell, 1922	Calcutta	Sewell (1922)
11.	<i>C. indicae</i> XVI Sewell, 1922	Calcutta	Sewell (1922)
12.	<i>C. indicae</i> XVIII Sewell, 1922	Calcutta	Sewell (1922)
13.	<i>C. indicae</i> XIX Sewell, 1922	Nilambur	Sewell (1922)
14.	<i>C. indicae</i> XXXIV Sewell, 1922	Wayanad	Sewell (1922)

1	2	3	4
15.	<i>C. indicae</i> XLVI Sewell, 1922	Mepadi, Wayanad	Sewell (1922)
16.	<i>C. indicae</i> XLVII Sewell, 1922	Wayanad	Sewell (1922)
17.	<i>C. indicae</i> LVI Sewell, 1922	Calcutta	Sewell (1922)
18.	<i>Cercaria patialensis</i> Soparkar, 1924 (= <i>Cercaria of Transversotrema patialense</i> (Soparkar, 1924) Cruz and Sathananthan, 1960)	Punjab, Nellur, Waltair	Soparkar (1924), Anantaraman (1948), Rao and Ganapati (1967)
19.	<i>C. quadriglandula</i> Premvati, 1953	Lucknow	Premvati (1953a)
20.	<i>C. magnacrestata</i> Premvati, 1953	Lucknow	Premvati (1953a)
21.	<i>C. crucita</i> Premvati, 1953	Lucknow	Premvati (1953b)
22.	<i>C. caudiglandula</i> Premvati, 1954	Pinjore, Chandigarh	Premvati (1954)
23.	<i>C. tuberculata</i> Premvati, 1954	Lucknow	Premvati (1954)
24.	<i>C. tuniforka</i> Premvati, 1954	Lucknow	Premvati (1954)
25.	<i>C. microcotyle</i> Peter, 1955	Madras	Peter (1955a)
26.	<i>C. multiplicata</i> Premvati, 1955	Lucknow	Premvati (1955)
27.	<i>C. reniforma</i> Premvati, 1956	Lucknow	Premvati (1956)
28.	<i>C. flavidusi</i> Premvati, 1956	Lucknow	Premvati (1956)
29.	<i>C. gomtiensis</i> Premvati, 1956	Lucknow	Premvati (1956)

1	2	3	4
30.	<i>C. bhimatalensis</i> Malaki and Singh, 1962	Pinjore	Malaki and Singh (1962)
31.	<i>C. naukuchiensis</i> Malaki and Singh, 1962	Pinjore, Chandigarh	Malaki and Singh (1962)
32.	<i>Cercaria</i> of <i>Philophthalmus nocturnus</i> Looss, 1907	Kondakarla	Murty (1966)
33.	<i>Cercaria diglandulata</i> Pandey, 1967	Lucknow	Pandey (1967b)
34.	<i>Cercaria</i> of <i>Neopronocephalus indicus</i> Mehra, 1932	Lucknow	Thapar (1968)
35.	<i>Cercaria kukrailensis</i> Thapar, 1969	Kukrail, Lucknow	Thapar (1969)
36.	<i>C. pinjorensis</i> Gupta and Taneja, 1969	Pinjore, Chandigarh	Gupta and Taneja (1969a)
37.	<i>Cercaria</i> (Monostome) sp. Gupta and Taneja, 1969	Pinjore, Chandigarh	Gupta and Taneja (1969a)
38.	<i>Cercaria chackai</i> Nadakal, Mohandas, Sunderaraman, 1969	Trivandrum	Nadakal <i>et al.</i> (1969)
39.	<i>C. soparkari</i> Pandey, 1971	Lucknow	Pandey (1971)
40.	<i>C. trioculata</i> Pandey, 1973	Lucknow	Pandey (1973b)
41.	<i>Cercaria</i> sp. III Kerala Mohandas, 1976	Palakkad, Ernakulam, Trivandrum	Mohandas (1976)
42.	<i>Cercariae indicae</i> LXVII Murty, 1976	Waltair	Murty (1976)
43.	<i>Cercaria guptai</i> Srivastava, 1977	Lucknow	Srivastava (1977)
44.	<i>Cercaria</i> of <i>Neopronocephalus triangularis</i> Mehra, 1932	Lucknow	Saxena (1977)

1	2	3	4
45.	<i>Cercaria buckleyi</i> Pandey and Agarwal, 1978	Lucknow	Pandey and Agarwal (1978)
46.	<i>C. lali</i> Pandey and Agarwal, 1978	Lucknow	Pandey and Agarwal (1978)
47.	<i>C. srivastava</i> Pandey and Agarwal, 1978	Lucknow	Pandey and Agarwal (1978)
48.	<i>C. martini</i> Pandey and Agarwal, 1978	Lucknow	Pandey and Agarwal (1978)
49.	<i>C. multiglandulata</i> Pandey and Agarwal, 1978	Lucknow	Pandey and Agarwal (1978)
50.	<i>C. chauhani</i> Pandey and Agarwal 1978	Lucknow	Pandey and Agarwal (1978)
51.	<i>C. nabhia</i> Gupta and Sharma, 1979	--	Gupta and Sharma (1979)
52.	<i>Cercaria</i> sp. II Kerala Mohandas, 1979	Calicut, Ernakulam, Palakkad	Mohandas (1979)
53.	<i>Cercariae indicae</i> LXXVI Sultana, 1980	--	Sultana (1980)
54.	<i>Cercaria</i> of <i>Haplorchoides vacha</i> sp. nov. Agrawal and Agrawal, 1981	--	Agrawal and Agrawal (1981)
55.	<i>Cercaria chauhani</i> Agrawal and Singh, 1981	Lucknow	Agrawal and Singh (1981)
56.	<i>Cercaria</i> of <i>Centrocestus formosanus</i> (Nishigori, 1924) Price, 1932	Andhra Pradesh	Ganapati and Rao (1983), Dhanumkumari <i>et al.</i> (1993a)
57.	<i>Cercaria johrii</i> Choubisa, 1985	Udaipur	Choubisa (1985)

1	2	3	4
58.	<i>Cercaria</i> of <i>Haplorchoides mehrai</i> Pandey and Shukla, 1976	Waltair	Shameem and Madhavi (1988)
59.	<i>Cercaria caudifina</i> Hyalij and Deoray, 1990	Nasik	Hyalij and Deoray (1990a)
60.	<i>Cercaria</i> sp. XVI Punjab Singh and Bali, 1990	Ludhiana	Singh and Bali (1990)
61.	<i>C. raipurensis</i> XI Ramachandrula and Agarwal 1990	Lucknow	Ramachandrula and Agarwal (1990)
62.	<i>C. visakhapatnamensis</i> 4 Dhanumkumari, Rao and Shyamasundari, 1990	Visakhapatnam	Dhanumkumari <i>et al.</i> (1990a)
63.	<i>C. visakhapatnamensis</i> 5 Dhanumkumari, Rao and Shyamasundari, 1990	Visakhapatnam	Dhanumkumari <i>et al.</i> (1990b)
64.	<i>Cercaria</i> sp. VII Bali and Singh, 1990	Punjab	Bali and Singh (1990)
65.	<i>C. jaikedia</i> Deoray and Hyalij, 1990	Nasik	Deoray and Hyalij (1990a)
66.	<i>C. viskhapatnamensis</i> I Dhanumkumari, Rao and Shyamasundari, 1991	Visakhapatnam	Dhanumkumari <i>et al.</i> (1991a)
67.	<i>Cercaria</i> of <i>Echinochasmus bagulai</i> Verma 1935	Visakhapatnam	Dhanumkumari <i>et al.</i> (1991b)
68.	<i>Cercaria udaipurensis</i> II Choubisa, 1992	Udaipur	Choubisa (1992)
69.	<i>C. machilipatnamensis</i> Dhanumkumari, Rao and Shyamasundari, 1993	Machilipatnam	Dhanumkumari <i>et al.</i> (1993a)

1	2	3	4
70.	<i>C. oesophagia</i> Hyalij and Deoray, 1995	Nasik	Hyalij and Deoray (1995)
71.	<i>C. spindale</i> Deoray and Hyalij, 1995	Nasik	Deoray and Hyalij (1995)
72.	Cercaria of <i>Orthetotrema monostomum</i> Macy and Basch, 1972	Mehadrigedda	Madhavi and Swarnakumari (1995)

Table 5. List of cercariae reported from *Paludomus* spp. in India

Sl. No.	Name	Locality	Reference
1.	<i>Cercariae indicae</i> XXXIII Sewell, 1922	Nilambur	Sewell (1922)
2.	<i>C. indicae</i> XXXVII Sewell, 1922	Wayanad	Sewell (1922)
3.	<i>C. indicae</i> XXXVIII Sewell, 1922 (= Cercaria of <i>Plagioporus panchax</i> n.sp.)	Wayanad	Sewell (1922)
4.	<i>C. indicae</i> XLV Sewell, 1922	Wayanad	Sewell (1922)
5.	<i>C. indicae</i> LVIII Sewell, 1922	Nilambur	Sewell (1922)

Table 6. List of cercariae reported from *Lymnaea* spp. in India

Sl. No.	Name	Locality	Reference
1	2	3	4
1.	Cercaria of <i>Schistosoma spindale</i> Montgomery, 1906 (= Cercaria of <i>S. spindalis</i> Liston and Soparkar, 1918)	Bombay	Liston and Soparkar (1918), Soparkar (1921a)
2.	<i>Cercaria bombayensis</i> No.8 Soparkar, 1921	Bombay	Soparkar (1921b)
3.	<i>C. bombayensis</i> No.19 Soparkar, 1921	Bombay	Soparkar (1921b)
4.	<i>Cercariae indicae</i> XVII Sewell, 1922	Bombay	Sewell (1922) Bhalerao (1943)
5.	<i>C. indicae</i> XXII Sewell, 1922	Calcutta	Sewell (1922)
6.	<i>C. indicae</i> XXIII Sewell, 1922	Calcutta Madras	Sewell (1922), Rao (1933c)
7.	<i>C. indicae</i> XXIV Sewell, 1922	Calcutta	Sewell (1922)
8.	<i>C. indicae</i> XXVIII Sewell, 1922	Bombay	Sewell (1922)
9.	<i>C. indicae</i> XXIX Sewell, 1922 (= Cercaria of <i>Fischoederius elongatus</i> (Poirer, 1833) Stiles and Goldberger, 1910)	Calcutta Madras	Sewell (1922), Rao and Ayyar (1932), Mukherjee (1966a)
10.	<i>C. indicae</i> XXX Sewell, 1922 (= Cercaria of <i>Schistosoma nasale</i> Rao, 1933)	Calcutta	Sewell (1922), Rao (1933a)
11.	<i>C. indicae</i> LIII Sewell, 1922	Wayanad	Sewell (1922)
12.	<i>Cercaria saundersi</i> Rao, 1932	Madras	Rao (1932a)
13.	<i>C. hurleyi</i> Rao, 1933	Madras	Rao (1933b)

1	2	3	4
14.	<i>C. pigmentosa</i> Bhalerao, 1933	Almora	Bhalerao (1933)
15.	<i>Cercariae indicae</i> X Sewell, 1922	Nizamsagar	Bhalerao (1943)
16.	<i>Cercaria insundhwessi</i> Porter, 1938	Arepally, Achampet	Bhalerao (1943)
17.	<i>Cercariae indicae</i> IX Sewell, 1922	Achampet, Ankol	Bhalerao (1943)
18.	<i>Cercaria mudaliari</i> Peter, 1955	Madras	Peter (1955a)
19.	<i>C. ramanujami</i> Peter, 1955	Madras	Peter (1955c)
20.	<i>Cercaria</i> of <i>Echinostoma revolutum</i> (Frollich, 1802) Dietz, 1909	Madras, Jaipur, Palakkad, Trivandrum	Peter (1955b), Patnaik and Ray (1966), Mohandas (1973a), Mukherjee (1992)
21.	<i>Cercaria rithaianensis</i> Singh, 1955	Allahabad	Singh (1955c)
22.	<i>C. soraonensis</i> Singh, 1955	Allahabad	Singh (1955a)
23.	<i>C. kumari</i> Singh, 1955	Allahabad	Singh (1955a)
24.	<i>C. biocellata</i> Singh, 1955	Allahabad	Singh (1955b)
25.	<i>C. stylata</i> Agarwal, 1956	Jabalpur	Agarwal (1956)
26.	<i>Cercaria</i> of <i>Clinostomum giganticum</i> Agarwal, 1959	Jabalpur	Agarwal (1959)
27.	<i>Cercaria</i> of <i>Clinostomum piscidium</i> Southwell and Prashad, 1918	Jabalpur, Lucknow	Singh (1959a), Pandey (1973c)
28.	<i>Cercaria</i> of <i>Echnoparyphum baquali</i> Jain, 1960	Ludhiana	Jain (1960a), Singh and Bali (1991)
29.	<i>Cercaria osmaniae</i> Simha, 1964	Hyderabad, Dadumajan, Chandigarh	Simha (1964), Gupta and Taneja (1968)

1	2	3	4
30.	<i>Cercaria (Gymnocephalous)</i> sp. Thapar and Tandon, 1952 (= <i>Cercaria</i> of <i>Fasciola gigantica</i> Cobbold, 1855)	Patiala	Thapar and Tandon (1952), Gupta and Taneja (1968)
31.	<i>C. ocellata</i> Lavalette St. George, 1855	Kharan, Chandigarh	Gupta and Taneja (1969b)
32.	<i>Cercaria (Lophocerca)</i> sp. inq. Gupta and Taneja, 1969	Patiala	Gupta and Taneja (1969b)
33.	<i>C. ghailae</i> Thapar 1969	Lucknow	Thapar (1969)
34.	<i>Cercariae indicae</i> XXV Sewell, 1922	Kukrail	Thapar (1969)
35.	<i>Cercaria limnaei</i> I Pandey and Srivastava, 1970	Lucknow	Pandey and Srivastava (1970)
36.	<i>C. limnaei</i> II Pandey and Srivastava, 1970	Lucknow	Pandey and Srivastava (1970)
37.	<i>Cercaria</i> of <i>Echinostoma malayanum</i> Leiper, 1911	Lucknow	Mohandas (1971b), Singh and Bali (1991)
38.	<i>Cercaria oviglandulata</i> Pandey, 1973	Lucknow	Pandey (1973a)
39.	<i>Cercaria</i> of <i>Echinoparyphium hymani</i> Singh, 1975	Durg	Singh (1975a)
40.	<i>Cercaria</i> of <i>Echinoparyphium lanceolatum</i> Singh, 1975	Durg	Singh (1975b)
41.	<i>Cercaria granulosa</i> Baugh, 1975	Lucknow	Baugh (1975)
42.	<i>Cercariae indicae</i> LXIII Murty, 1975	Kondakarla	Murty (1975a)
43.	<i>C. indica</i> LXIV Murty, 1975	Kondakarla	Murty (1975a)
44.	<i>Cercaria</i> of <i>Echinoparyphium vitellocompactum</i> Singh, 1976	Durg	Singh (1976)

1	2	3	4
45.	<i>Cercaria dharmtallensis</i> Mukherjee and Ghosh, 1977	Calcutta	Mukherjee and Ghosh (1977)
46.	<i>C. dietzi</i> Singh, 1977	Durg	Singh (1977)
47.	<i>C. itoli</i> Pandey and Agarwal, 1977	Lucknow	Pandey and Agarwal (1977)
48.	<i>C. chillavanensis</i> Agrawal and Sharma, 1980	Lucknow	Agrawal and Sharma (1980)
49.	<i>C. khargapurensis</i> Jain and Sharma, 1981	Khargapur	Jain and Sharma (1981)
50.	<i>Cercaria</i> sp. VI Kerala Mohandas, 1981	Trivandrum	Mohandas (1981)
51.	<i>Cercaria</i> sp. VII Kerala Mohandas, 1981	Trichur, Palakkad, Ernakulam	Mohandas (1981)
52.	<i>Cercaria</i> sp. VIII Kerala Mohandas, 1981	Palakkad, Trivandrum	Mohandas (1981)
53.	<i>Cercaria raipurensis</i> VII Ramachandrula and Agarwal, 1985	Raipur	Ramachandrula and Agarwal (1985)
54.	<i>C. raipurensis</i> VIII Ramachandrula and Agarwal, 1985	Raipur	Ramachandrula and Agarwal (1985)
55.	<i>Cercaria</i> of <i>Haematolechus</i> <i>almorai</i> Pande, 1937	Waltair	Madhavi and Shameem (1986)
56.	<i>Cercaria hanumanthai</i> Pandey and Singh, 1986	Lucknow	Pandey and Singh (1986)
57.	<i>C. baughi</i> Pandey and Singh, 1986	Lucknow Unnao	Pandey and Singh (1986)
58.	<i>C. raipurensis</i> I Ramachandrula and Agarwal, 1987	Raipur	Ramachandrula and Agarwal (1987)

1	2	3	4
59.	<i>C. raipurensis</i> III Ramachandrula and Agarwal, 1987	Raipur	Ramachandrula and Agarwal (1987)
60.	<i>C. godavaria</i> Deoray and Hyalij, 1990	Nasik	Deoray and Hyalij (1990b)
61.	<i>Cercaria</i> sp. X. Punjab Singh and Bali, 1991	Ludhiana	Singh and Bali (1991)
62.	<i>Cercaria</i> sp. XI Punjab Singh and Bali, 1991	Ludhiana	Singh and Bali (1991)
63.	<i>Cercaria</i> of <i>Echinostoma</i> <i>cinetorchis</i> Ando and Ozaki, 1923	Gurdaspur	Singh and Bali (1991)
64.	<i>Cercaria</i> of <i>Echinoparyphium</i> <i>flexum</i> (Linton, 1892) Dietz, 1910	Ludhiana	Singh and Bali (1991)
65.	<i>Cercaria</i> of <i>Paryphostomum</i> <i>giganticum</i> Rai and Agarwal, 1961	Kannur, Kozhikode	Nambiar and Janardanan (2001)

Table 7. List of cercariae reported from *Indoplanorbis* spp. in India

Sl. No.	Name	Locality	Reference
1	2	3	4
1.	Cercaria of <i>Schistosoma spindalis</i> Liston and Soparkar, 1918	Bombay	Soparkar (1921a)
2.	<i>Cercaria bombayensis</i> No.9 Soparkar, 1921	Andheri	Soparkar (1921a)
3.	<i>C. bombayensis</i> No.13 Soparkar, 1921	Andheri	Soparkar (1921b)
4.	<i>Cercariae indicae</i> I Sewell, 1922	Russa Road	Sewell (1922)
5.	<i>C. indicae</i> II Sewell, 1922 (= Cercaria of <i>Diplostomum ketupanense</i> Vidyarthi, 1937)	Calcutta, Wayanad, Kannur	Sewell (1922), Roopa and Janardanan (2001)
6.	<i>C. indicae</i> IX Sewell, 1922	Calcutta, Sultan Battery, Wayanad	Sewell (1922)
7.	<i>C. indicae</i> X Sewell, 1922	Calcutta, Wayanad	Sewell (1922)
8.	<i>C. indicae</i> XII Sewell, 1922	Calcutta	Sewell (1922)
9.	<i>C. indicae</i> XVII Sewell, 1922	Calcutta, Madras	Sewell (1922), Peter (1955a)
10.	<i>C. indicae</i> XX Sewell, 1922	Calcutta	Sewell (1922)
11.	<i>C. indicae</i> XXI Sewell, 1922	Calcutta	Sewell (1922)
12.	<i>C. indicae</i> XXV Sewell, 1922	Ganjan	Sewell (1922)
13.	<i>C. indicae</i> XXVI Sewell, 1922 (= Cercaria of <i>Cotylophoron indicum</i> Stile and Goldberger, 1910)	Wayanad, Nilambur, Calcutta, Madras, Palakkad	Sewell (1922), Rao (1932c), Peter (1956b), Mukherjee (1960), Mohandas (1976)

1	2	3	4
14.	<i>C. indicae</i> XXVII Sewell, 1922	Calcutta	Sewell (1922)
15.	<i>C. indicae</i> XXX Sewell, 1922 (= <i>Cercaria</i> of <i>Schistosoma nasalis</i> Rao, 1933)	Calcutta	Sewell (1922), Rao (1933a)
16.	<i>C. indicae</i> XXXVI Sewell, 1922	Wayanad	Sewell (1922)
17.	<i>C. indicae</i> XLVIII Sewell, 1922	Wayanad	Sewell (1922)
18.	<i>C. indicae</i> LIII Sewell, 1922	Calcutta	Sewell (1922)
19.	<i>Cercaria mehrai</i> Faruqui, 1930	Lucknow, Allahabad, Madras, Ropar	Faruqui (1930), Peter (1955b), Jain (1958, 1960b), Singh and Bali (1991)
20.	<i>C. kylasami</i> Rao, 1932	Madras	Rao (1932b)
21.	<i>Cercaria</i> of <i>Cotylophoron cotylophorum</i> Stiles and Goldberger, 1910	Lucknow	Srivastava (1938)
22.	<i>Cercaria fraseri</i> Buckley, 1939 (= <i>Cercaria</i> of <i>Gastrodiscus secundus</i> (Looss, 1907))	Madras	Buckley (1939), Peter and Mudaliar (1948), Anantaraman & Balasubramaniam (1949), Mukherjee (1986)
23.	<i>Cercaria</i> of <i>Schistosoma indicum</i> Montgomery, 1906	Allahabad	Srivastava and Dutt (1951)
24.	<i>Cercaria (Clinostome) hunterii</i> Singh, 1952	Ahimanpur	Singh (1952a)

1	2	3	4
25.	<i>Cercaria of Plasmiorchis orientalis</i> Mehra, 1934	Allahabad	Singh (1952b)
26.	<i>Cercaria anuri</i> Singh, 1952	Hundia	Singh (1952c)
27.	<i>C. ahimanpurensis</i> Singh, 1953	Ahimanpur	Singh (1953)
28.	<i>C. longicaudata</i> Singh, 1953	Alfred park	Singh (1953)
29.	<i>C. hornifurca</i> Singh, 1953	Sobatiabagh	Singh (1953)
30.	<i>C. sphericauda</i> Singh, 1953	Allahabad	Singh (1953)
31.	<i>C. delhupurensis</i> Singh, 1953	Allahabad	Singh (1953)
32.	<i>Cercaria of Pseudodiscus collinsi</i> (Cobbold, 1975) Stiles and Goldberger, 1910	Bareilly	Peter and Srivastava (1954, 1960a)
33.	<i>Cercaria kanihari</i> Singh, 1955	Ahimanpur	Singh (1955c)
34.	<i>C. nairi</i> Peter, 1955	Madras	Peter (1955b)
35.	<i>C. bareillyi</i> Peter and Srivastava, 1955	Barielly	Peter and Srivastava (1955)
36.	<i>C. indoplanorbis</i> Peter and Srivastava, 1955	Barielly	Peter and Srivastava (1955, 1960b)
37.	<i>C. robertsoni</i> Patki, 1956	Jabalpur	Patki (1956)
38.	<i>Cercaria lewertii</i> Singh, 1957 (= <i>Cercaria of Srivastavaia lewertii</i> (Singh, 1970) Mukherjee, 1986)	Lucknow	Singh (1957) Mukherjee (1986)
39.	<i>C. mainpurensis</i> Mukherjee, 1963	Manipuri	Pandey (1965)
40.	<i>C. dalibaghensis</i> Pandey, 1967	Lucknow	Pandey (1967)
41.	<i>C. tetraglandulata</i> Srivastava, 1968	Lucknow	Srivastava (1968)

1	2	3	4
42.	<i>C. exusta</i> Thapar, 1969	Rae Barelli, Lucknow	Thapar (1969)
43.	<i>C. chauhani</i> Pandey and Jain, 1971	Lucknow, Agra	Pandey and Jain (1971)
44.	<i>Cercaria</i> of <i>Echinostoma malayanum</i> Leiper, 1911	Trivandrum	Mohandas (1971)
45.	<i>Cercaria megaglandulata</i> Agrawal, 1974	Durg	Agrawal (1974)
46.	<i>Cercariae indicae</i> LXV Murty, 1975	Kondakarla	Murty (1975a)
47.	<i>C. triglandulata</i> Baugh, 1975	Lucknow	Baugh (1975)
48.	<i>Cercaria</i> sp. I. Kerala Mohandas, 1976	Palakkad, Trivandrum	Mohandas (1976)
49.	<i>C. chinahatensis</i> Srivastava, 1978	Lucknow	Srivastava (1978)
50.	<i>C. tandani</i> Pandey and Singh, 1982	Sultanpur	Pandey and Singh (1982)
51.	<i>C. nariandapurensis</i> Pandey and Singh, 1984	Sultanpur	Pandey and Singh (1984)
52.	<i>C. nawabganjensis</i> Pandey and Singh, 1984	Unnao	Pandey and Singh (1984)
53.	<i>C. tewarii</i> Choubisa and Sharma, 1984	Udaipur	Choubisa and Sharma (1984)
54.	<i>C. raipurensis</i> VIII Ramachandrula and Agarwal, 1985	Raipur	Ramachandrula and Agarwal (1985)
55.	<i>C. ramalingami</i> Pandey and Singh, 1986	Meerut, Unnao	Pandey and Singh (1986)

1	2	3	4
56.	<i>C. raipurensis</i> I Ramachandrula and Agarwal, 1985	Raipur	Ramachandrula and Agarwal (1987)
57.	<i>C. raipurensis</i> II Ramachandrula and Agarwal, 1987	Raipur	Ramachandrula and Agarwal (1987)
58.	<i>C. raipurensis</i> IV Ramachandrula and Agarwal, 1987	Raipur	Ramachandrula and Agarwal (1987)
59.	<i>Cercaria</i> sp. IX Punjab Bali and Singh, 1991	Sangrur, Ludhiana	Bali and Singh (1991)
60.	<i>Cercaria</i> of <i>Protechinostoma</i> <i>macronisertulatum</i> Beaver, 1943	Ropar	Singh and Bali (1991)
61.	<i>Cercaria banglabazarensis</i> I sp. Srivastava and Solomon, 1992	Lucknow	Srivastava and Solomon (1992)
62.	<i>C. banglabazarensis</i> II sp. Srivastava and Solomon, 1992	Lucknow	Srivastava and Solomon (1992)
63.	<i>C. prayagpurensis</i> Srivastava, 1992	Prayagpur, Lucknow	Srivastava (1992)
64.	<i>C. satpalai</i> Agarwal, Singh and Maurya, 1993	Varanasi	Agarwal <i>et al.</i> (1993)
65.	<i>C. mowayiansis</i> Agarwal, Singh and Maurya, 1993	Varanasi	Agarwal <i>et al.</i> (1993)
66.	<i>C. jaunpurensis</i> Maurya, Yadav, Prajapati and Agrawal, 1996	Ranjitpur, Jaunpur	Maurya <i>et al.</i> (1996)
67.	<i>Cercaria</i> of <i>Euclinostomum</i> <i>heterostomum</i> (Rudolphi, 1809)	Visakhapatnam	Jhansilakshmibai and Madhavi (1997)
68.	<i>Cercaria baughi</i> Pandey and Panwar, 2000	Meerut	Pandey and Panwar (2000)

Table 8. List of cercariae reported from *Gyraulus* spp. in India

Sl. No.	Name	Locality	Reference
1.	<i>Cercariae indicae</i> IX Sewell, 1922	Calcutta	Sewell (1922)
2.	<i>C. indicae</i> XXIX Sewell, 1922 (= Cercaria of <i>Fischoederius elongatus</i> (Poirier, 1883) Stiles and Goldberger, 1910)	Calcutta, Madras	Sewell (1922) Peter (1955b)
3.	<i>C. indicae</i> XII Sewell, 1922	Calcutta	Sewell (1922)
4.	<i>Cercaria gyraulusi</i> Peter and Srivastava, 1955 (= cercaria of <i>Gigantocotyle explanatum</i> (Creplin, 1847) Nasmark, 1937)	Lucknow, Bareilly	Peter and Srivastava (1955), Singh (1958)
5.	<i>C. chungathi</i> Peter and Srivastava, 1955 (= Cercaria of <i>Gastrothylax crumerifer</i> (Creplin, 1847) Otto, 1896)	Rithora, Lucknow	Peter and Srivastava (1955)
6.	Cercaria of <i>Oliveria indica</i> Thapar and Sinha, 1945	Lucknow	Thapar (1961)
7.	<i>Cercaria fursolensis</i> Singh and Malaki, 1963	Bhowali, Kumaun	Singh and Malaki (1963)
8.	<i>C. rithorensis</i> Mukherjee, 1963	Mathurapur	Mukherjee (1963)
9.	<i>C. megacuda</i> Baugh, 1975	Lucknow	Baugh (1975)
10.	<i>C. kondakarali</i> I Shameem and Madhavi, 1984	Koodakarala, Waltair	Shameem and Madhavi (1984)
11.	Cercaria of <i>Posthodiplostomum grayii</i> (Verma, 1936)	Visakhapatnam	Madhavi and Rukmini (1997)

MATERIALS AND METHODS

Vasandakumar M.V. “Studies on the larval trematodes (digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India” Thesis. Department of Zoology , University of Calicut, 2002

MATERIALS AND METHODS

Collection and Maintenance of Snail hosts

Freshwater snails, used during the course of this investigation, were collected from rivulets, streams, ponds, paddy fields and irrigation canals in Kasaragod, Kannur, Wayanad, Kozhikode, Malappuram and Palakkad districts of Malabar, Kerala during a 71 month period, from February 1996 through December 2001. Collections were made using hand nets or scoop nets and by hand picking. Altogether 10 species of snails belonging to 8 genera were collected during the course of this investigation. A list of the snails examined, with their localities, period of collection and number examined, is presented in Table 9.

The snails collected from their natural habitats were brought alive to the laboratory in polythene bags and maintained in groups of 5-10 in beakers containing well-water. The mouths of the beakers were covered with fine-meshed nets to prevent the escape of snails and to provide good aeration. The water in the containers was changed daily and the snails were exposed to sunlight at convenience. The snails were fed occasionally with grated potato and water plants. The water in the beakers was checked daily for the presence of cercariae, and on detection of cercariae the snails were transferred to separate beakers and maintained individually so that the snails which shed cercariae could be identified.

Table 9. List of host specimens examined

Classification and name of host	No. of host specimens examined	Locality of collection	Period of collection
1	2	3	4
Phylum : Mollusca			
Class : Gastropoda			
Subclass : Prosobranchia			
Order : Mesogastropoda			
Family : Viviparidae			
Subfamily : Bellamyinae			
<i>Bellamyia bengalensis</i> (Lamarck, 1882)	269	Athiyamboor, Neeleswaram and Thrikaripur in Kasaragod district; Kunhimangalam, Payangadi Pappinassery, Thalap, Koodali and Mangattidam in Kannur district; Feroke in Kozhikode district; Kizhuparamba and Pallikkal Bazar in Malappuram district.	April - May 1996; June - July 1997; September 1998; June to September 1999; July and December 2000.
<i>Bellamyia dissimilis</i> (Mueller, 1774)	628	Athiyamboor, Neeleswaram and Thrikaripur in Kasaragod district; Cherukunnu, Pappinassery, Pallikunnu, Kavinmoola Iriveri, Koodali, Chala, Andalur, Palayad, Munderi and Eachur in Kannur district; Chorode, Kundayithode, Feroke, Ramanattukara and Nallur in Kozhikode district; Kizhuparamba, Idimuzhikkal, Pallikal Bazar, Azhinjilam, Iruvetty and Olippuram Kadavu in Malappuram district	June 1996; January 1997; January to October 1998; June 1999; May to November 2000; January to June 2001, November - December 2001.

1	2	3	4
Family : Pilidae			
<i>Pila virens</i> (Lamarck, 1822)	889	Padanakkad and Neeleswaram in Kasaragod district; Pappinassery, Cherukunnu, Pallikunnu, Azhikkal, Munderi, Iriveri, Valyannur, Peralassery, Andalur, Palayad, Mangattidam and Patteri in Kannur district; Kundayithode, Ramanattukara, Nallur, Kadalundi and Vadakara in Kozhikode district; Iruvetty, Nilambur, Idimuzhikkal, Olippuram Kadavu, Kizhuparamba and Thalappara in Malappuram district	February and November 1996; February 1998; October - November 1998; January - February 2000; June to December 2000; January to October 2001.
Family : Bithynidae			
Subfamily : Bithyninae			
<i>Digoniostoma pulchella</i> (Benson, 1836)	36	Malampuzha in Palakkad district.	August - September 2000.
Family : Thiaridae			
Subfamily : Thiarinae			
<i>Thiara scabra</i> (Mueller, 1774)	178	Theerthamkara and Neeleswaram in Kasaragod district; Thazhe Chovva, Avera, Koodali, Munderi, Anjarakandy and Kanhileri in Kannur district; Sultan Bathery in Wayanad district; Kizhuparamba, Karulayi and Nilambur in Malappuram district	January - February 1997. August - September 1998; August to November 2000; February to April 2001.

1	2	3	4
<i>Thiara tuberculata</i> (Mueller, 1774)	1161	Kanhangad, Athiyamboor, Padanakkad, Theerthamkara and Neeleswaram in Kasaragod district; Cherukunnu, Thazhe Chovva, Avera, Kuruva, Munderi, Iritty, Peravoor, Kanhileri, Koodali and Anjarakandy in Kannur district; Sultan Bathery in Wayanad district, Feroke and Kadalundi in Kozhikode district; Kizhuparamba, Thalappara, Azhinjilam, Chaliyar and Nilambur in Malappuram district; Malampuzha in Palakkad district.	February - March 1996; August to November 1996; June to September 1998; March 2000; August to December 2000; January to November 2001.
Subfamily : Paludominae			
<i>Paludomus transchauricus</i> (Gmelin, 1771)	1615	Padanakkad, Athiyamboor and Theerthamkara in Kasaragod district; Pallikunnu, Avera, Thazhe Chovva, Anjarakandy, Pappinassery, Iritty, Kanhileri, Patteri and Kolleri in Kannur district; Vattakundu in Wayanad district; Kundaythode and Nallur in Kozhikode district; Kizhuparamba, Nilambur, Karulayi and Thalappara in Malappuram district; Malampuzha in Palakkad district.	October to December 1996; July-August 1997; January 1998; June to September 1998; April to December 2000; April to December 2001.

1	2	3	4
Subclass : Pulmonata			
Order: Basommatophora			
Family: Lymnaeidae	1943	Kanhangad and Theerthamkara in Kasaragod district; Cherukunnu, Pappinassery, Thazhe Chovva, Peralassery, Azhikode, Andalur, Palayad, Kanhileri, Patteri and Iritty in Kannur district; Payyoli, Vadakara, Feroke and Nallur in Kozhikode district, Anayarangadi, Kizhuparamba, Azhinjilam, Nilambur, Iruvetty and Idimuzhikal in Malappuram district; Malampuzha in Palakkad district.	July to October 1996; July and November 1997; January to March 1998; November 1998; April to December 2000; January to December 2001.
<i>Lymnaea luteola</i> Lamarck, 1822			
Family : Planorbidae			
Subfamily : Bulininae			
<i>Indoplanorbis exustus</i> (Deshayes, 1834)	2545	Neeleswaram, Athiyamboor, Padanakkad, Thrikaripur and Theerthamkara in Kasaragod district; Payyanur, Cherukunnu, Kalliassery, Pappinassery, Pallikunnu, Azhikkal, Muzhappilangad, Thazhe Chovva, Andalur, Palayad, Eachur, Mundayad, Peralassery, Thalavil, Chala, Manantheri, Mangattidam, Kanhileri, Patteri and Peravoor in Kannur district; Vadakara, Payyoli, Feroke, Ramanattukara, Kadalundi	September to November 1996; July to September 1997; January 1998; July to December 1998; May to December 2000; January to December 2001.

1	2	3	4
Subfamily: Planorbinae		and Nallur in Kozhikode district; Kizhuparamba, Idimuzhikkal, Pallikkal Bazar, Nedungottumedu, Iruvetty, Thalappara, Azhinjilam, Nilambur, Karulayi, Anayarangadi and Olippuram Kadavu in Malappuram district; Malampuzha in Palakkad district.	
<i>Gyraulus convexiusculus</i> (Hutton, 1849)	83	Andalur in Kannur district; Malampuzha in Palakkad district	August 2000; January to March 2001.

Study of cercaria

Naturally emerged fresh cercariae were subjected to detailed studies on their morphology and behaviour. Swimming behaviour of the cercariae was studied by transferring them into a petridish containing water and observing with a powerful hand lens or under a binocular dissecting microscope. The cercariae were then transferred on to a glass slide, stained with vital stains like neutral red, Nile blue sulphate or toluidine blue, covered with a No.1 cover glass, and observed under a Leitz Diaplan phase-contrast microscope to study their internal organization. The use of stain mixture containing equal proportions of neutral red, Nile blue sulphate and toluidine blue made the structural details of cercariae more clear. Collar spines and cuticular spines of cercariae were studied by staining them with dilute Lugol's iodine solution (0.01%). The spines were visible in the form shining, light diffracting formations on the yellowish background of the body. The cercariae were stained with 0.1% Nile blue sulphate in saline, refrigerated (14°C) for 30 min, and observed under the microscope to study the excretory system. Mounting the cercariae in 1% urea solution also facilitated the study of excretory system. Genital primordia were studied by staining the cercariae with acetocarmine. For permanent preparations, the cercariae were fixed in 10% formalin, stained with alum carmine, dehydrated in ethanol grades, cleared in xylol and mounted in DPX. Measurements were taken from 10% formalin-fixed cercariae.

Study of intramolluscan stages

Infected snails which shed cercariae were later dissected and their body tissues transferred into a petri-dish containing 0.65% saline. Intramolluscan stages of development of cecariae can be detected with naked eyes as yellow or white mottlings. Pieces of infected tissues were then smeared on slides, covered with cover glass, and examined for developing stages of sporocysts, rediae and cercariae. These smears were observed under the phase-contrast microscope with or without vital staining. The smears were made permanent by fixing in hot 10% formalin, and staining in alum carmine.

Studies on life cycles

Search for second intermediate/definitive hosts

Natural habitats of infected snails were searched for second intermediate hosts of trematodes. The host snails, other snails of the habitat, stonefly naiads, mayfly and dragonfly nymphs, prawns, crabs, fishes, tadpoles and frogs were collected from the water bodies where infected snails were present. These hosts were brought alive to the laboratory, immediately dissected and examined for the presence of metacercarial infections.

Natural habitats of snails and second intermediate hosts were explored for definitive hosts of flukes. Also the areas surrounding these habitats were searched

for the definitive hosts. The suspected definitive hosts collected were fishes like *Aplocheilus panchax*, *Rasbora daniconius* and *Danio aequipinnatus*, the pond heron, *Ardeola grayii*, the little cormorant, *Phalacrocorax niger* and the cattle egret, *Bubulcus ibis*. The wild-caught hosts were taken to the laboratory, dissected and examined for adult flukes.

Study of metacercariae and adults

Metacercariae and adults, when present, were carefully removed from infected tissues and lumen of internal organs of hosts, and collected in 0.75% saline taken in petri-dishes. Metacercarial cysts were transferred on to a glass slide containing saline and observed under a stereo-zoom dissecting microscope for structural details of cysts and enclosed larvae. The larva was taken out of the cyst either by rupturing the cyst wall with the help of fine needles or by mounting cysts under cover glass and applying gentle pressure over it by fine needles. Excysted larvae and adults were studied under the phase-contrast microscope with or without staining by neutral red or methylene blue. Permanent whole mounts of excysted larvae and adults were prepared after fixing them in 10% formalin under slight cover glass pressure, and then staining with alum carmine, following Cantwell (1981). The flukes were subsequently identified to get an idea of the various species of digeneans occurring in the study area.

Experimental infection studies

With the background knowledge obtained from the study on larval and adult digeneans, experiments were conducted in the laboratory to trace the life cycles using clean natural hosts and experimental animals. Life cycles were successfully established for two species of trematodes, *Plagioporus panchax* n. sp. and *Petasiger variospinosus* (Odhner, 1910). Partial life cycle from cercaria to fully developed metacercaria was traced for a species of *Microparyphium*.

Cercariae from natural infections were used in experimental infection studies. The cercariae of *P. panchax* shed by *Paludomus transchauricus* were exposed to clean stonefly naiads to develop their metacercariae. The infective metacercariae from natural infections or those developed in the laboratory were fed to infection-free natural definitive hosts such as *Aplocheilus panchax*, *Rasbora daniconius* and *Danio aequipinnatus* to obtain adults. The stonefly naiads and fishes were dissected at various intervals post-exposure and development of metacercariae and adults were followed.

In the case of *Petasiger variospinosus*, the 27-spined echinostome cercariae shed by naturally infected *Indoplanorbis exustus* were used to develop their metacercariae in *Rasbora daniconius*, *Puntius parrah* and *Esomus barbatus*. In order to obtain adults, the infected fishes were fed to the little cormorant, *Phalacrocorax niger* free from parasites as determined by faecal investigations for

the presence of eggs of *P. variospinus*. The fed hosts were then maintained in the laboratory, dissected at various intervals, and the flukes at different stages of development were recovered. Flukes were also recovered from naturally infected *P. niger*.

Eggs were obtained by tearing the uterus of gravid flukes and incubated at room temperature (26-28°C) to study the development and release of miracidia. Miracidial morphology was studied in live specimens. Silver impregnation technique of Lynch (1933) was employed to study the number and arrangement of epidermal plates.

The echinostome cercariae emerged from *Pila virens* were used to develop their metacercariae in *Danio aequipinnatus*, *Aplocheilus lineatus*, *Labeo rohita* and *Macropodus cupanus*. The fishes exposed to cercariae were examined at intervals to observe the development of metacercariae. The metacercariae were identified as belonging to the genus *Microparyphium*. Studies on further development of the metacercaria were conducted in one-day-old chicks and white rats; these were unsuccessful.

Measurements and sketches

Measurements were taken with the aid of a calibrated ocular micrometer. All measurements are given in micrometres (μm); the range is followed by the mean values in parentheses. Description of each stage in the digenean life cycle is

based on measurements of a minimum of 15 specimens. Sketches were drawn with the aid of a prism type camera lucida and details added free hand from observations made on live specimens.

RESULTS

Vasandakumar M.V. “Studies on the larval trematodes (digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India” Thesis. Department of Zoology , University of Calicut, 2002

RESULTS

Cercaria sp. I Malabar n. sp.

Infection by this cercaria was found in the planorbid snail, *Indoplanorbis exustus*, collected from paddy fields and ponds at Pallikunnu and Andalur in Kannur district during October - November 1998 and May 2001. Out of 163 snails collected from these localities 13 were found infected.

Cercariae emerged from snails throughout day time, with maximum emergence in the morning. They exhibited swimming movements typical of strigeids. During resting, the cercaria remained suspended in the water column with the body directed downwards.

Description: (Figs. 1-2a, Table 10)

Brevifurcate, apharyngeate, ocellate, distome cercaria. Body elongate, narrow anteriorly, spinose, 178-310 (251) long, 69-129 (97) wide. Tail stem spinose, attached ventrally, 419-749 (575) x 49-86 (72) in dimensions. Each furca with a dorso-ventral fin fold extending its whole length and four to six linear rows of spines; measured 168-217 (200) x 33-99 (50).

Anterior organ pyriform, 62-83 (74) x 42-69 (53) in size, with eight to ten rows of prominent spines in the anterior half. The organ possesses a pair of club-

shaped cephalic glands with well-developed nuclei in the anterior two-third and four pairs of nucleated cells in the posterior part. Acetabulum post-equatorial protrusible, circular, 29-43 (36) in diameter, with its aperture bordered by a circle of spines. A pair of deeply pigmented eyespots, 10-15 (12) in diameter, situated at anterior two-fifth of body.

Mouth subterminal. Oesophagus long, passes through the anterior organ, and ends blindly with slight dilation at the level of eyespots. Penetration glands 5 pairs, lateral to acetabulum; anterior 2 pairs with coarsely granular and the remaining 3 pairs with finely granular contents. Ducts of penetration glands on each side form a bundle, and open individually through minute pores, capped with hollow spines. A horse-shoe-shaped structure, filled with finely granular contents located behind penetration glands; its function not known. Genital primordium represented by a compact mass of small, round cells between acetabulum and horse-shoe-shaped structure.

Excretory bladder 'V' or 'Y' shaped, with six pairs of flame cells in the body, and one pair near the tail base. Flame cell formula $2 [(2+1) + (2+1) + (1)] = 14$. Caudal excretory duct runs through the middle of tail stem, then bifurcates, passes through each furca, and opens out at furcal tip.

100 μ m

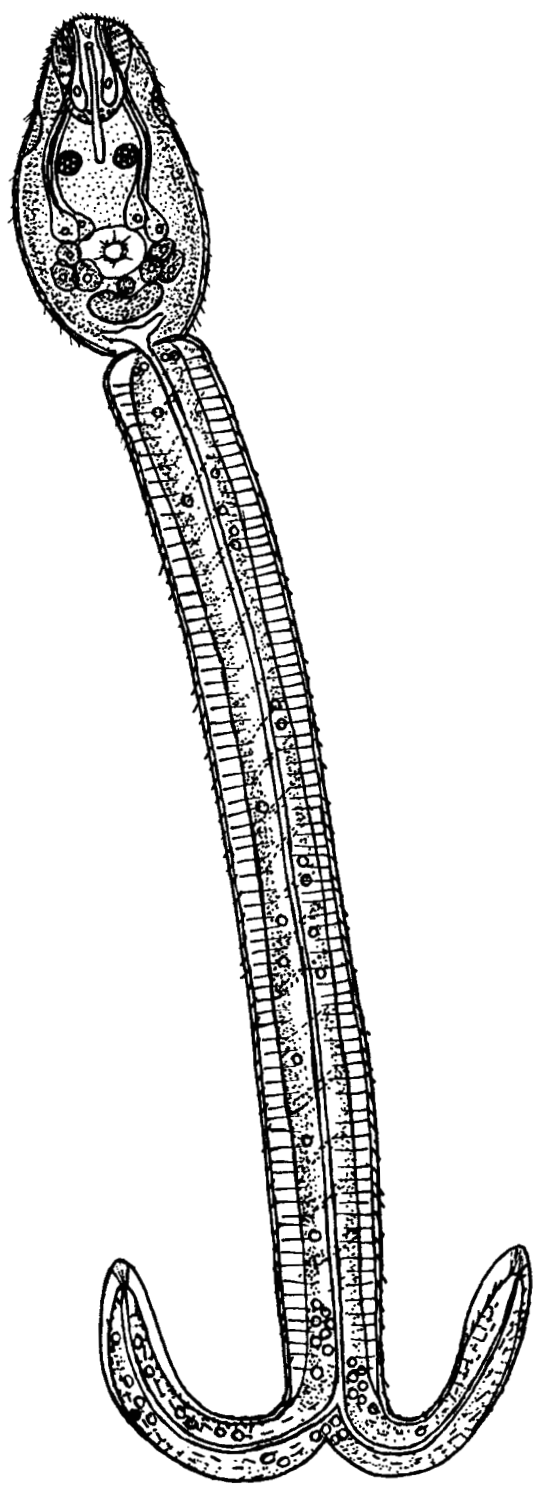


Fig. 1 : *Cercaria* sp. I Malabar n. sp.

Table 10. Measurements of *Cercaria* sp. I Malabar n. sp.

Characters	Range	Mean
Body	178 - 310 x 69 - 129	251 x 97
Tail	419 - 749 x 49 - 86	575 x 72
Furca	168 - 271 x 33 - 99	200 x 50
Anterior Organ	62 - 83 x 42 - 69	74 x 53
Acetabulum	29 - 43	36
Eyespot	10 - 15	12
Flame cell formula	$2[(2+1)+(2+1)+(1)] = 14$	

Sporocyst: (Fig. 2b)

Sporocysts oval, saccular, 158-238 (191) x 138 – 208 (162) in size and filled with 4-12 fully developed cercariae and several germ balls at different stages of development.

- Snail host : *Indoplanorbis exustus* (Deshayes)
- Site of infection : Hepatopancreas
- Locality : Pallikunnu and Andalur in Kannur district (Kerala)
- Period of collection : October-November 1998; May 2001.
- Prevalence : Thirteen of 2545 (0.51%) snails examined were infected.

Remarks:

The present brevifurcate, ocellate cercaria is comparable to *Cercaria bombayensis* no. 13 Soparkar, 1921, *Cercariae indicae* XXV Sewell, 1922 and *Cercaria wardi* Miller, 1926 in having furcal finfold and cephalic gland. But it differs from *C. bombayensis* no. 13 in the number of cephalic glands, and the number and pattern of flame cells in the body is different from that of the present species. *C. wardi* having median head gland, developed caeca, two groups of

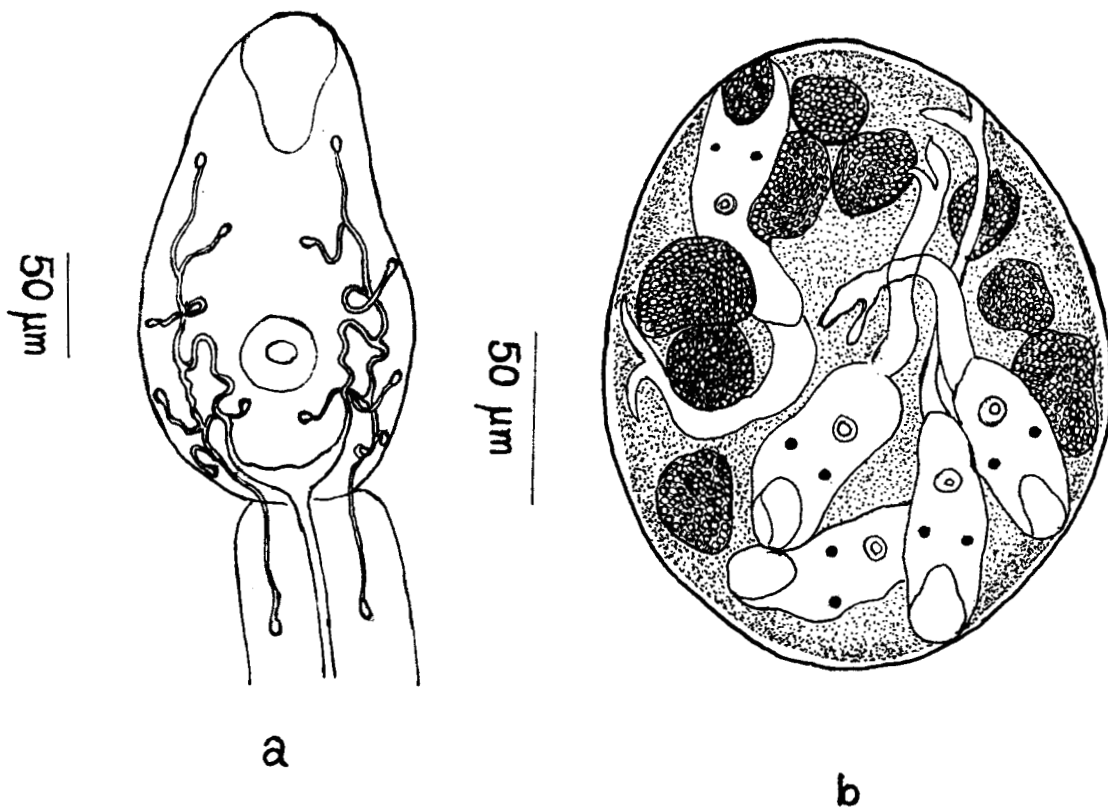


Fig. 2 : *Cercaria* sp. I Malabar n. sp.
a. Excretory system; b. Sporocyst

penetration glands with 4 and 6 in each, and 5 pairs of flame cells in body is distinct from the present species.

In view of the reasons stated, I am convinced that the present cercaria is a new species, and is reported here as *Cercaria* sp. I Malabar n. sp.

***Cercaria* sp. II Malabar n. sp.**

The cercaria was recovered from 4 of 45 *Indoplanorbis exustus* collected from Kizhuparamba in Malappuram district of Kerala, during December 2000. Snails collected from all other localities were negative for this infection.

The cercariae emerged from infected snails during early morning hours, and remained uniformly distributed in the water. Cercariae exhibited alternate ascending and descending swimming movements with short intervals of rest. During swimming the larva directed its tail foremost with the furca diverged maximally. Occasionally the larva directed its head forward and moved actively. The quick vibrating movements of the larva gave it an appearance of the figure '8'.

Description: (Figs. 3-4a, Table 11)

Brevifurcate, pharyngeate, distome cercaria. Body elongate, 148-275 (200) long and 82-140 (120) wide. Anterior organ pear-shaped, 26-65 (41) x 26-60 (40) in size, bears 10-14 rows of closely set, prominent spines at its anterior half, spines

in rest of the body sparsely distributed. Three pairs of sensory hairs on lateral margin of body. Tail attached postero-ventrally; tail stem aspinose with 4 pairs of sensory hairs, and lateral fin folds which extend the entire length of tail stem. Fin folds maximally wide at the mid level. Tail stem measured 171-382 (228) long, 60-100 (80) wide, without finfolds, and 125-185 (147) wide with finfolds. Furca spinose, with 4 pairs of sensory hairs; 125-231 (183) long, 33-69 (46) wide. Acetabulum post-equatorial, 19-33 (27) in diameter, with two alternating rows of spines.

Mouth subterminal. Prepharynx absent. Pharynx well developed, globular, muscular, 13-20 (17) in diameter. Oesophagus narrow, moderately long, 6-29 (17), bifurcates at pre-acetabular zone. Caeca long, extending to posterior end; each caecum divisible into 3 compartments. Penetration glands 4 pairs, intercaecal, encircling acetabulum; each gland with round nucleus and coarsely granular contents. Gland ducts of each side form a bundle, run anteriorly and open out near the tip. Genital primordium represented by a round mass of cells situated posterior to acetabulum. Excretory bladder 'Y' shaped, at posterior end of body. Flame cell formula $2[(2+2) + (2+2) + (2)] = 20$, with 2 pairs of flame cell in the tail stem. Caudal excretory duct passes through the tail stem, bifurcates at its distal end, run into each furca and opens out mid-furcally.

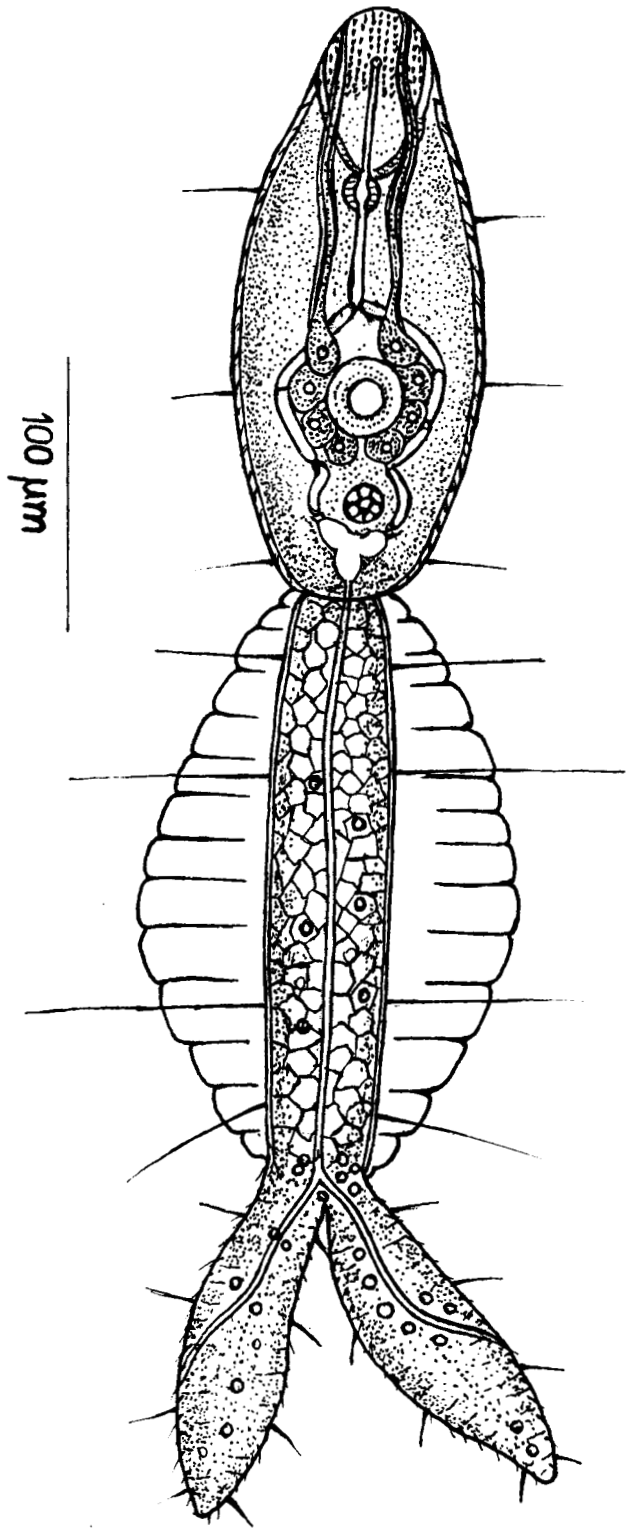


Fig. 3 : *Cercaria* sp. II Malabar n. sp.

Table 11. Measurements of *Cercaria* sp. II Malabar n. sp.

Characters	Range	Mean
Body	148 - 275 x 82 - 140	200 x 120
Tail	171 - 382 x 60 - 100	228 x 80
Furca	125 - 231 x 33 - 69	183 x 46
Anterior organ	26 - 65 x 26 - 60	41 x 40
Acetabulum	19 - 33	27
Pharynx	13 - 20	17
Oesophagus	6 - 29	17
Flame cell formula	$2[(2+2)+(2+2)+(2)] = 20$	

Sporocyst: (Fig. 4b)

Sporoysts were recovered from the hepatopancreas of *Indoplanorbis exustus*. Sporocysts elongate, reddish-yellow, 780-2932 (1916) x 124-592 (215) in size; enclose 28 to 86 cercariae and numerous germ balls at various stages of development

- Snail host : *Indoplanorbis exustus* (Deshayes)
- Site of infection : Hepatopancreas
- Locality : Kizhuparamba in Malappuram district (Kerala)
- Period of collection : December 2000
- Prevalence : Four of 2545 (0.16%) snails examined were infected.

Remarks:

The present cercaria is characterised by the presence of a peculiar finfold extending the whole length of tail. This type of finfold has not been observed in any of the freshwater furcocercous cercaria reported till date. However, it needs comparison with *Cercaria stephensi* Brooks, 1943, *C. lessoni* Johnston and Bewckwith, 1947, *C. sphaericauda* Singh, 1953 and cercaria of *Strigea sphaerula* (Rud., 1803) Mathias, 1925 (by Odening, 1967) in the number and disposition of

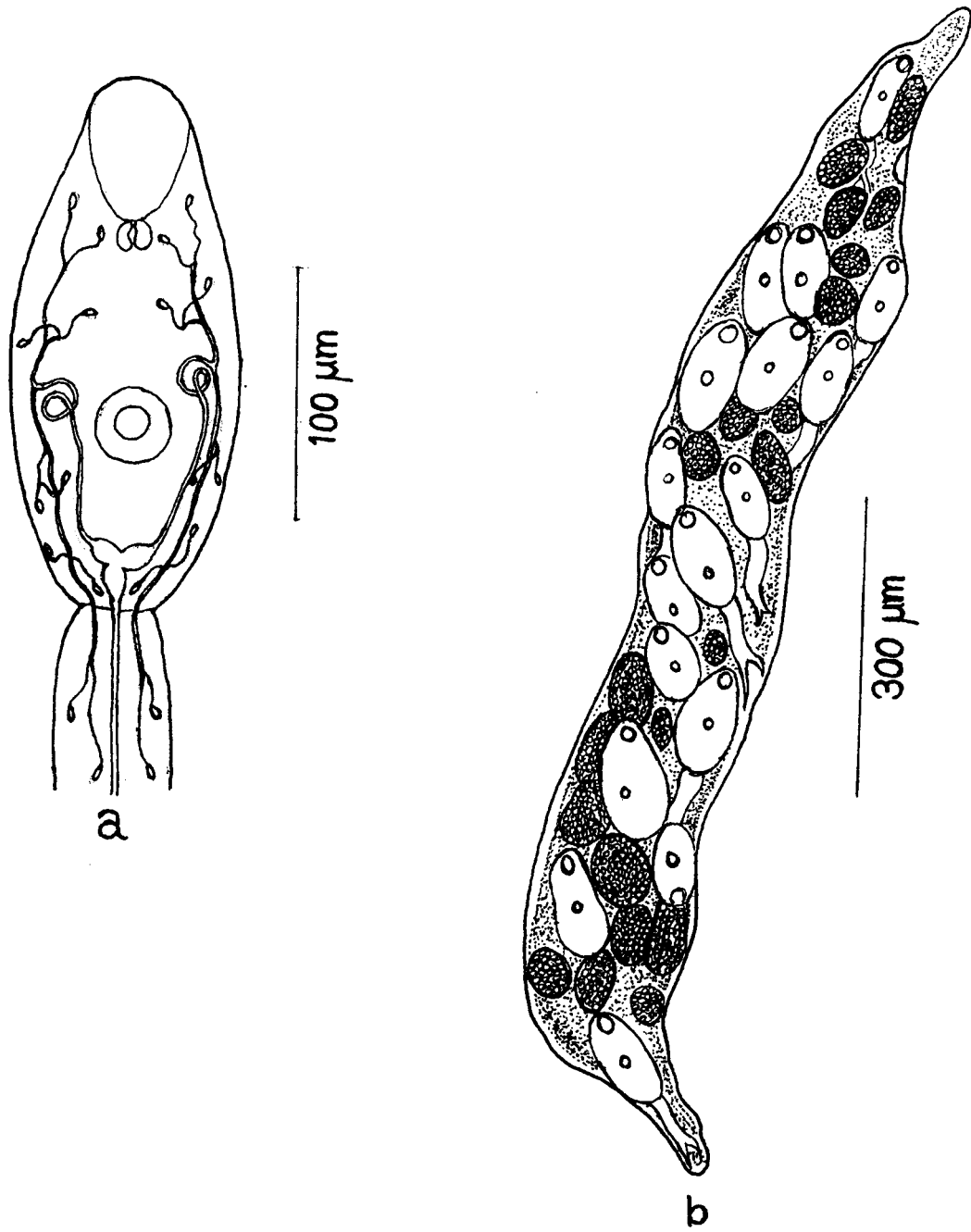


Fig. 4 : *Cercaria* sp. II Malabar n. sp.
a. Excretory system; b. Sporocyst

penetration glands. But it is different from all these cercariae in having a characteristic caudal finfold, in the nature of intestinal caeca, pattern of excretory system and in the absence of eyespots.

As the present cercaria cannot be identified with any other furcocercous cercariae reported earlier, it is considered as new and the name *Cercaria* sp. II Malabar n.sp. is proposed for it.

Cercaria of Transversotrema patialense
(Soparkar, 1924) Cruz and Sathananthan, 1960

(Fig. 5)

Host	:	<i>Thiara tuberculata</i> (Mueller)
Locality	:	Kuruva in Kannur district; Kizhuparamba in Malappuram district.
Period of collection	:	September 1998; March 2001.
Prevalence	:	Five of 1161 (0.43%) snails examined were infected.

Natural infections by the adult trematode were found in *Rasbora daniconius* collected from the above localities.

100 μ m

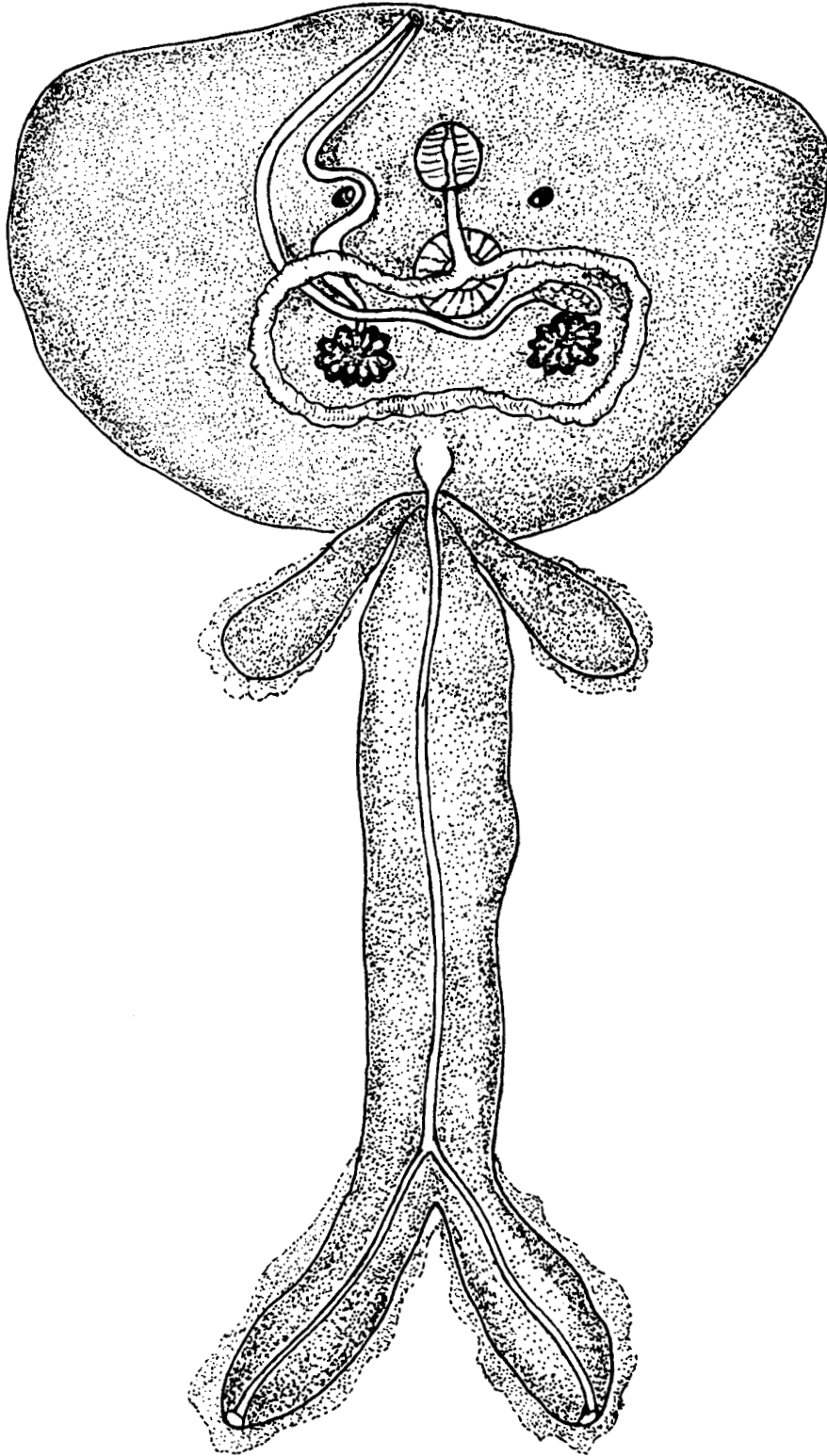


Fig. 5 : Cercaria of *Transversotrema patialense*
(Soparkar, 1924) Cruz and Sathananthan, 1960

Remarks:

Characters of the present cercaria agreed fully with that of *Cercaria patialensis*, given by Soparkar (1924) from *Melanoides tuberculatus* in Punjab. Subsequently, this cercaria was reported from the same snail species in Nellore by Anantaraman (1948), in Waltair by Rao and Ganapati (1967), in Ceylon by Crusz (1956) and in Malaysia by Betterton (1979), and from *M. anomala* in the Belgian Congo by Brien (1954).

Crusz and Sathananthan (1960) reported the metacercaria of *C. patialensis* from *Macropodus cupanus* in Ceylon and designated it *Transversotrema patialense*. Crusz *et al.* (1964) followed the development of *C. patialensis* in *Macropodus cupanus*, *Ophiocephalus punctatus* and *Thilapia mossambica* and preferred to call the stages recovered from fishes as "mature metacercariae." Rao and Ganapati (1967) also gave a detailed account of the cercaria of this species from *M. tuberculatus*, and adults from *Esomus danricus*, *Panchax panchax* and *Catla catla*. Further, they suggested that there may not be a metacercarial stage in the life cycle of *Transversotrema* spp. Subsequently, Murty and Rao (1968), Madhavi (1980a) and Rekharani and Madhavi (1985a) recorded adults from different fishes. In 1994, Madhavi and Jhansilakshmibai described its miracidium.

Two more species of transversotrematid cercariae were described from India, namely, *C. chackai* Nadakal *et al.*, 1969 from *M. tuberculata* and *M. scabra*

and *C. soparkari* Pandey, 1971 from *M. tuberculata*. Nadakal *et al.* (1969) differentiated *C. chackai* from *C. patialensis* by differences in body size and features of excretory system. Pandey (1971) differentiated *C. soparkari* from *C. patialensis* by the arrangement of excretory canals. Pande and Shukla (1972) described the adult of *C. soparkari* and named it *T. soparkari*. Mohandas (1973b) obtained the adults of *C. chackai* experimentally in fishes, named it *T. chackai* and stated that although the adult structures of different species are identical, there is divergence in their larval characters. But the two cercarial forms differed only slightly from *C. patialensis* in morphological features. Therefore, it appears reasonable to conclude that these two species may represent morphological variations of *T. patialense*.

***Cercaria* sp. III Malabar n. sp.**

Infection by this cercaria was encountered in *Lymnaea luteola* collected from paddy fields and rivulets of Feroke in Kozhikode district. Out of 1943 snails examined 12 were found infected by this cercaria showing an overall prevalence of 0.62%.

The cercariae emerged in large numbers in the morning hours. They exhibited characteristic swimming activity, with the tail forward in all directions, and the movements started and ended in jerks. During resting, the cercaria

remained suspended in the water column with the body hanging downward, tail stem turned upward and the furcae extended to the full. The cercaria performed leech-like movements on a glass slide with a thin film of water or at the bottom of the container.

Description: (Figs. 6-7a, Table 12)

Longifurcate, pharyngeate, ocellate, distome cercaria Body oval, 130-187 (161) long, 35-70 (51) wide, spinose; spines conspicuous at anterior region. Two pairs of short sensory hairs present, one at the level of eyespots and the other below the acetabulum. The tail stem aspinose, with ten pairs of sensory hairs, one pair at proximal end and the other pairs at the posterior half; measured 161-189 (174) x 28-43 (36). Six pairs of caudal bodies with well developed nuclei arranged on either side of the caudal excretory duct. Furcae 170-224 (209) long, 14-32 (24) wide, spinose, with two pairs of small setae on each furca.

Anterior organ pear-shaped, protrusible; 30-53 (40) x 28-39 (33) in size. Acetabulum round, just post-equatorial, 35-42(39) in diameter, provided with 3 rows of spines around its aperture. A pair of unpigmented eyespots, each with four units, present at the level of anterior margin of acetabulum.

Mouth subterminal; prepharynx short, 7-12 (10) in length. Pharynx small, spherical, 8-12 (10) in diameter; oesophagus, 11-24 (22) long. Caeca 40-75 (65) long, extending to the level of posterior margin of acetabulum; each caecum

constricted into three compartments. Penetration gland 4 pairs, post acetabular; each gland with coarsely granular contents and large nucleus. The gland ducts on each side form a bundle, run anteriorly, and open near the mouth. Genital primordium represented by a mass of cells located ventrally, posterior to the acetabulum.

Excretory bladder V or Y-shaped, thin walled. Flame cells 14 pairs with one pair in tail. Flame cell formula, $2[(2+2) + (2)+(1)] = 14$. Caudal excretory duct runs through the tail stem, then bifurcates and open out through midfurcal pores.

Sporocyst: (Fig. 7b)

Sporocysts thread-like, ash-coloured, 1232-4851 (2986) long, 77-123 (94) wide; contain numerous cercariae and germ balls at various stages of development.

- Snail host : *Lymnaea luteola* Lamarck
- Site of infection : Hepatopancreas
- Locality : Feroke in Kozhikode district (Kerala)
- Period of collection : November 1998; May 2001
- Prevalence : Twelve of 1943 (0.62%) snails examined were infected.

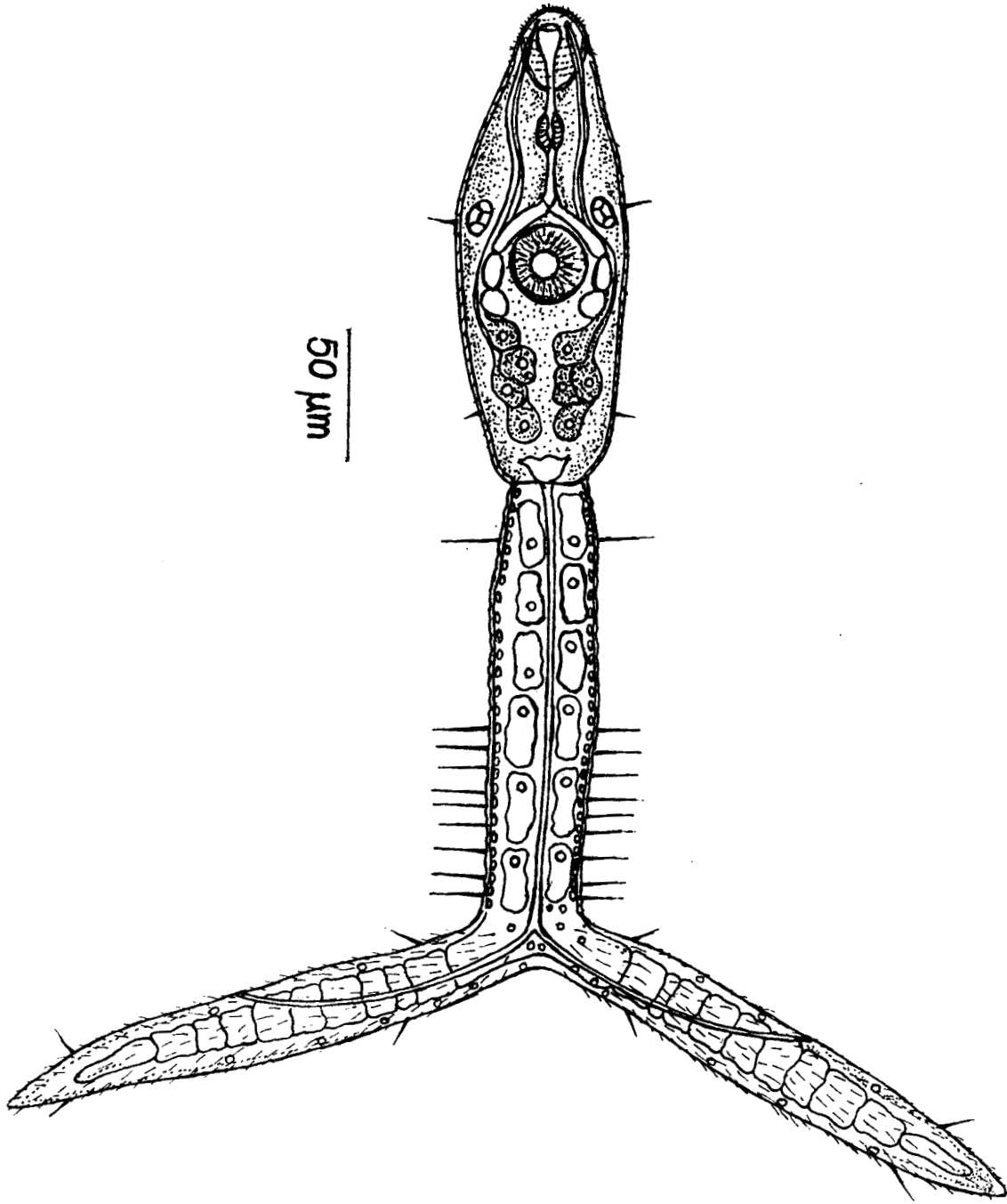


Fig. 6 : *Cercaria* sp. III Malabar n. sp.

Table 12. Measurements of *Cercaria* sp. III Malabar n. sp.

Characters	Range	Mean
Body	130 - 187 x 35 - 70	161 x 51
Tail	161 - 189 x 28 - 43	174 x 36
Furca	170 - 224 x 14 - 32	209 x 24
Anterior organ	30 - 53 x 28 - 39	40 x 33
Acetabulum	35 - 42	39
Prepharynx	7 - 12	10
Pharynx	8 - 12	10
Oesophagus	11 - 24	22
Caeca	40 - 75	65
Flame cell formula	$2[(2+2)+(2)+(1)] = 14$	

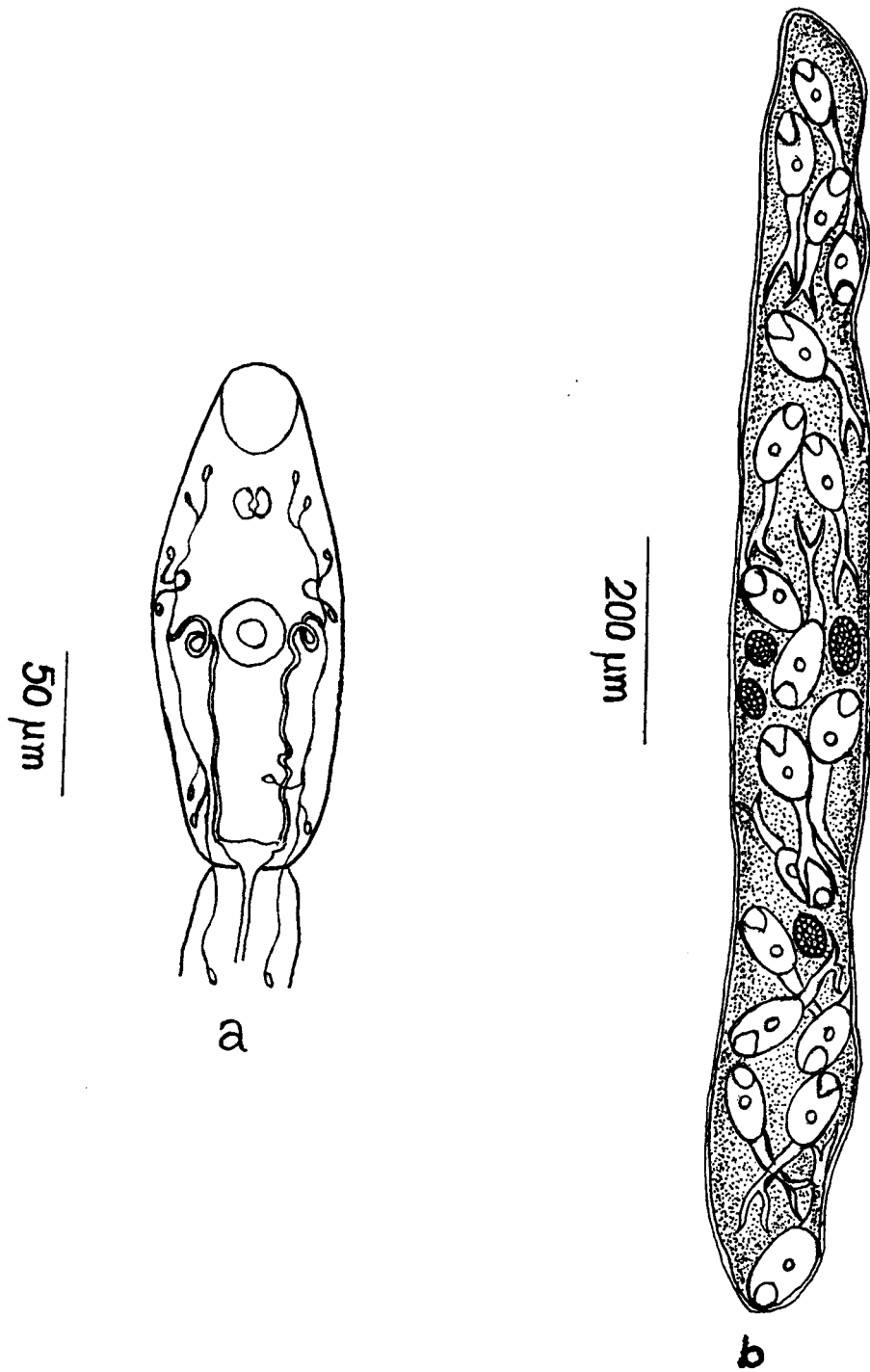


Fig. 7 : *Cercaria* sp. III Malabar n. sp.
a. Excretory system; b. Sporocyst

Remarks:

Among the known furcocercous cercariae, the present form infecting *Lymnaea luteola* comes close to *Cercaria gracillima* Faust, 1917, *C. longiremis* Wesenberg-Lund, 1934, Cercaria of *Strigea elegans* Chandler et Rausch, 1947 (by Pearson, 1959), *Furcocercaria* sp. 3 Odening, 1962, Cercaria of *Strigea falconis palumbi* Viborg, 1975 and of *Strigea strigis* (Schrank, 1788) (by Odening, 1967) and *Furcocercaria* sp. VIII Ostrowski de Nuenez *et al.*, 1996 in having four pairs of post-acetabular penetration glands and caudal bodies in tail, and in the absence of anterior and / or posterior excretory commissures.

C. gracillima is distinctly different from the present form in the absence of pharynx. *C. longiremis*, cercaria of *S. elegans* and *Furcocercaria* sp. 3 differ from the species under discussion in the absence of eyespots, extent of caeca beyond the acetabular level and in the number and arrangement of spines and sensory hairs. In the linear arrangement of penetration glands, number of flame cells, extent of caeca and in the number of caudal bodies and sensory hairs in the tail of cercariae of *S. falconis palumbi* and *S. strigis* and *Furcocercaria* sp. VIII are different from the present form.

In view of the reasons stated above, the present cercaria is considered as a new species and named *Cercaria* sp. III Malabar n. sp.

Cercaria sp. IV Malabar n. sp.

The planorbid snails, *Indoplanorbis exustus*, were found infected by the cercaria during October - November, 2000. The infected snails were collected from Payyoli in Kozhikode district. Nine of 2545 snails examined were infected.

Cercariae emerged from snails during evening hours and were found uniformly distributed in water. Cercariae exhibited characteristic swimming behaviour with prolonged resting periods alternating with short spells of swimming activity. During resting phase, the larva remained suspended in the water column with the body directed downwards, tail stem upwards and furcae diverged to maximum extent. Leech-like movements were not observed.

Description: (Figs. 8-9a, Table 13)

Longifurcate, pharyngeate, ocellate, distome cercaria. Body elongate-oval, with a pear-shaped anterior organ; measured 132-177 (149) long, 36-60 (48) wide. Tail stem aspinose, 145-198 (176) long and 33-43 (38) wide, provided with 14 pairs of long sensory hairs. Furcae 178-231 (209) x 19-23 (22) in size, provided with longitudinal rows of spines; two comparatively short sensory hairs on each furca.

Anterior organ highly muscular, measured 33-40 (38) x 26-40 (32), with 8 to 11 rows of prominent, backwardly directed spines in the anterior half. Two cephalic glands on either side of oral cavity in anterior organ. Eleven pre-oral

spines in three transverse rows; 3 spines in the row nearest the mouth and 4 each in the second and third rows. Two bands of spines encircling the body present at the posterior end of anterior organs; spines in between the bands sparsely distributed. Rest of the body devoid of spines. Acetabulum rudimentary, located at the end of anterior two third of body. Two non-pigmented eyespots at mid region of body.

Mouth subterminal, prepharynx short; pharynx spherical, 9-13 (11) in diameter. Oesophagus and caeca not discernible. Penetration glands 3 pairs, post-equatorial, surrounding acetabulum; each gland with finely granular contents and large nucleus. Gland ducts of each side form a bundle, take a forward course, pass over anterior organ and open at the anterior margin through 3 pores, guarded by spines. Cystogenous glands numerous, beneath the tegument. Genital primordium represented by a conical mass of cells located just in front of excretory bladder.

Excretory bladder oval, at posterior end of body. Flame cell formula $2[(2)+(2+2+2)+(2)] = 20$, with posterior two pairs in anterior third of tail stem. Caudal excretory duct runs through the tail stem, bifurcates at its distal end into two branches, run into the furcae and open out mid-furcally.

Sporocyst: (Fig. 9b)

Sporocysts reddish brown, elongated, thread-like, 1794-6068 (3590) x 93 - 203 (134) in size; contain 8-9 well-developed cercariae, a few developing cercariae and germ balls at different stages of development.

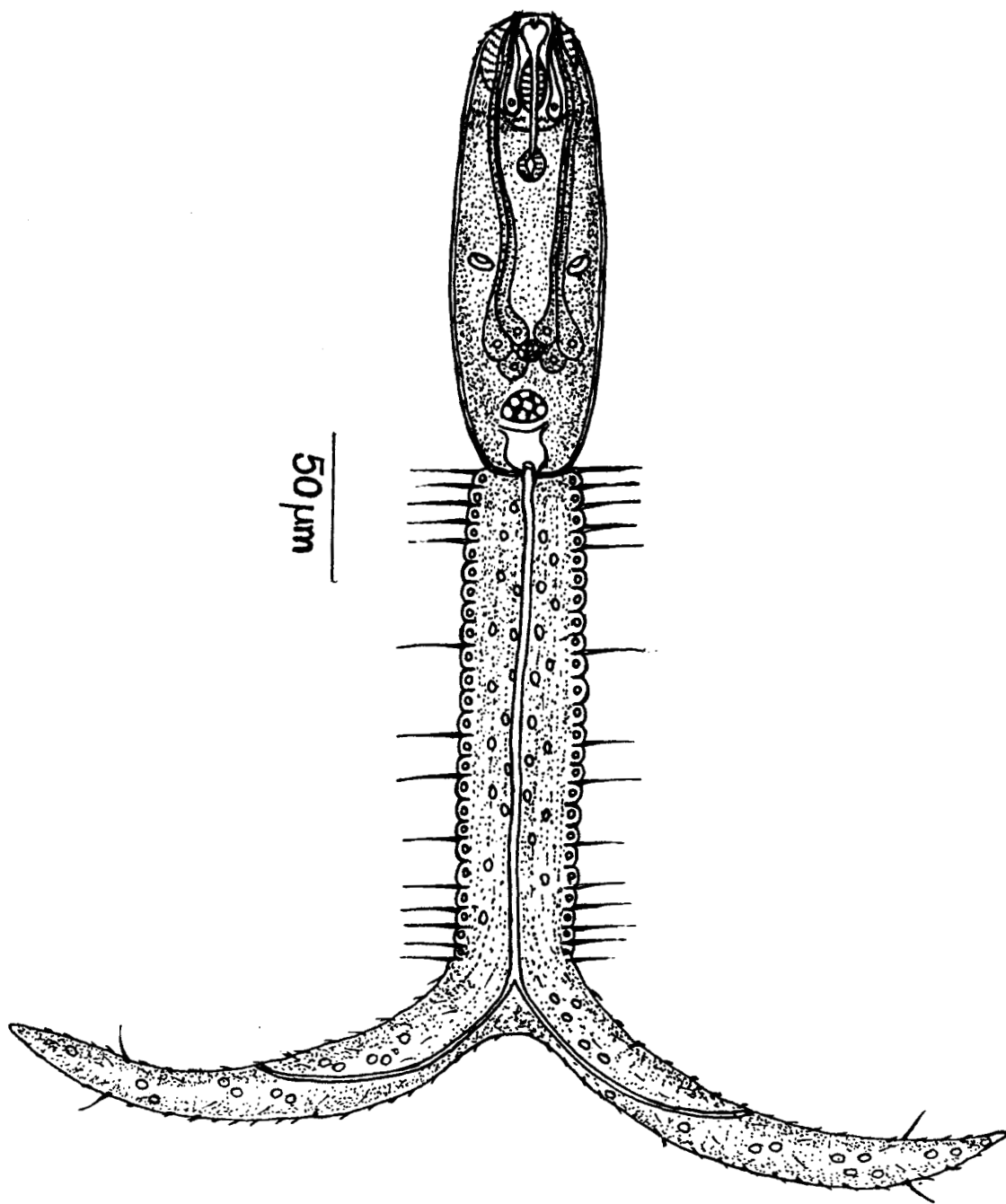


Fig. 8 : *Cercaria* sp. IV Malabar n. sp.

Table 13. Measurements of *Cercaria* sp. IV Malabar n. sp.

Characters	Range	Mean
Body	132 - 177 x 36 - 60	149 x 48
Tail	145 - 198 x 33- 43	176 x 38
Furca	178 - 231 x 19-23	209 x 22
Anterior organ	33 - 40 x 26 - 40	38 x 32
Pharynx	9 - 13	11
Flame cell formula	$2[(2)+(2+2+2)+(2)] = 20$	

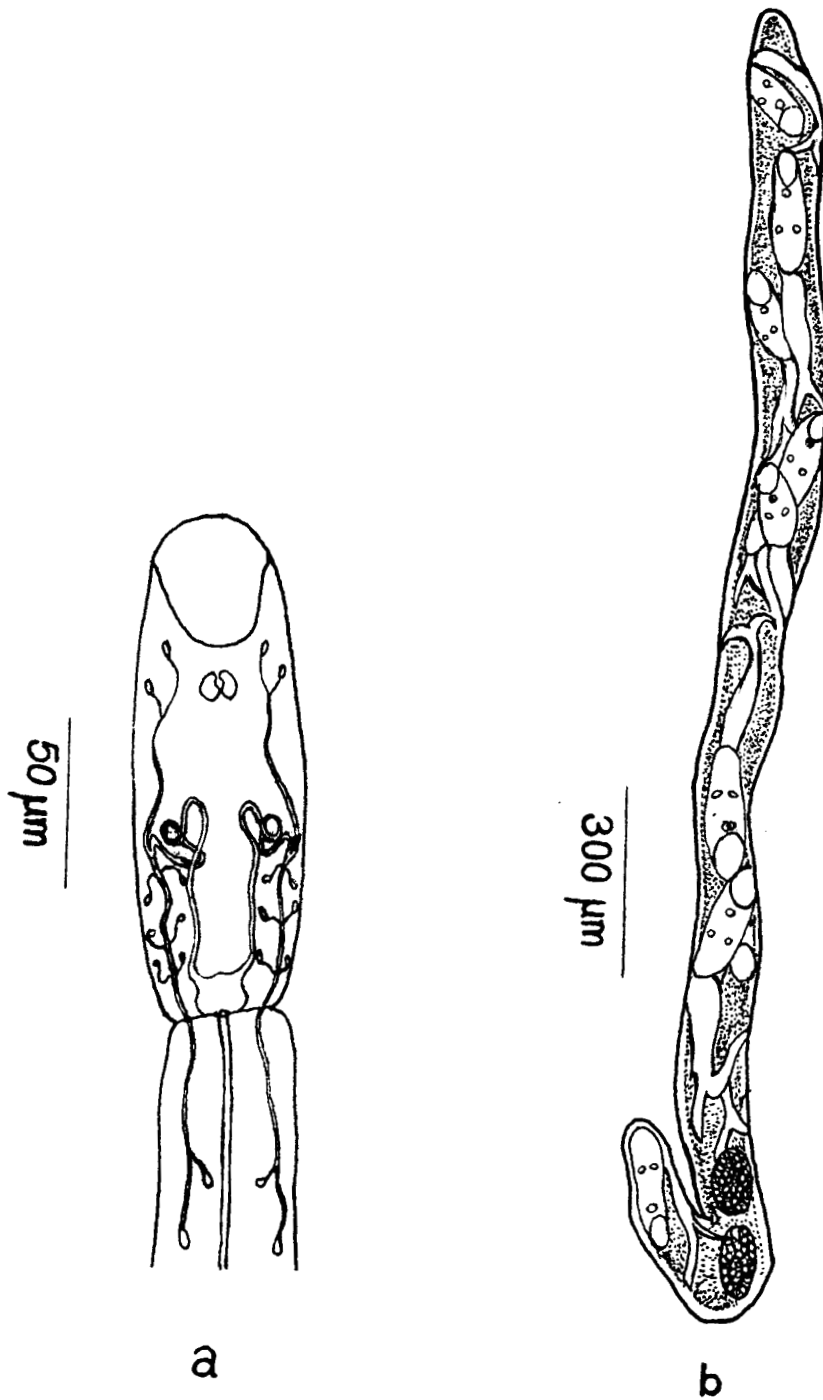


Fig. 9 : *Cercaria* sp. IV Malabar n. sp.
a. Excretory system; b. Sporocyst

- Snail host : *Indoplanorbis exustus* (Deshayes)
- Site of infection : Hepatopancreas
- Locality : Payyoli in Kozhikode district (Kerala)
- Period of collection : October-November 2000
- Prevalence : Nine of 2545 (0.35%) snails examined were infected.

Remarks:

Of the various longifurcate cercariae with eyespots and 3 pairs of penetration glands, the present form needs comparison with *Cercaria multicellulata* Miller, 1924, *C. physae* Cort and Brooks, 1928, *C. louisiana* Miller, 1935, *Cercaria* of *Posthodiplostomum minimum* (Mac Callum, 1921) (by Miller, 1954), *C. kaniharri* Singh, 1955, *C. dalibaghensis* Pandey, 1967, *C. leyteensis* no. 29 Ito and Blas, 1977, *C. emirati* V Isamil and Arif, 1991 and *Cercaria* of *Diplostomum grayii* (Verma, 1936) (by Madhavi and Rukmini, 1997). But in the presence of pharynx, unpigmented eyespots, arrangement of penetration glands and number of flame cells, it is distinctly different from *C. physae*, *C. louisiana*, *Cercaria* of *P. minimum*, *C. leyteensis* no. 29 and *C. emirati* V. *C. dalibaghensis* deviates from the present cercaria in having prominent acetabulum and cellular caeca, in the arrangement of penetration glands and in the number of flame cells. Presence of pigmented eyespots in *C. kaniharri* and cercaria of *D. grayii* makes them distinct

from the present form which has only unpigmented eyespots. In the arrangement of penetration glands also these cercariae show significant differences. Cercaria of *D. grayii* differs further from the present cercaria in having developed caeca and finfolds on furcae.

Since the present cercaria is distinctly different from the other related cercariae, it appears reasonable to consider the present form as a new species and is named *Cercaria* sp. IV Malabar n. sp.

***Cercaria of Diplostomum ketupanense* Vidyarthi, 1937**

(Fig. 10)

Host	:	<i>Indoplanorbis exustus</i> (Deshayes)
Locality	:	Kizhuparamba, Malappuram district
Period of collection	:	November – December 2000; January 2001.
Prevalence	:	Eleven of 2545 (0.43%) snails examined were infected.

Metacercariae of *D. ketupanense* were observed in the muscle tissues beneath the scales of *Rasbora daniconius* collected from Kizhuparamba in Malappuram district. Prevalence of infection was very high.

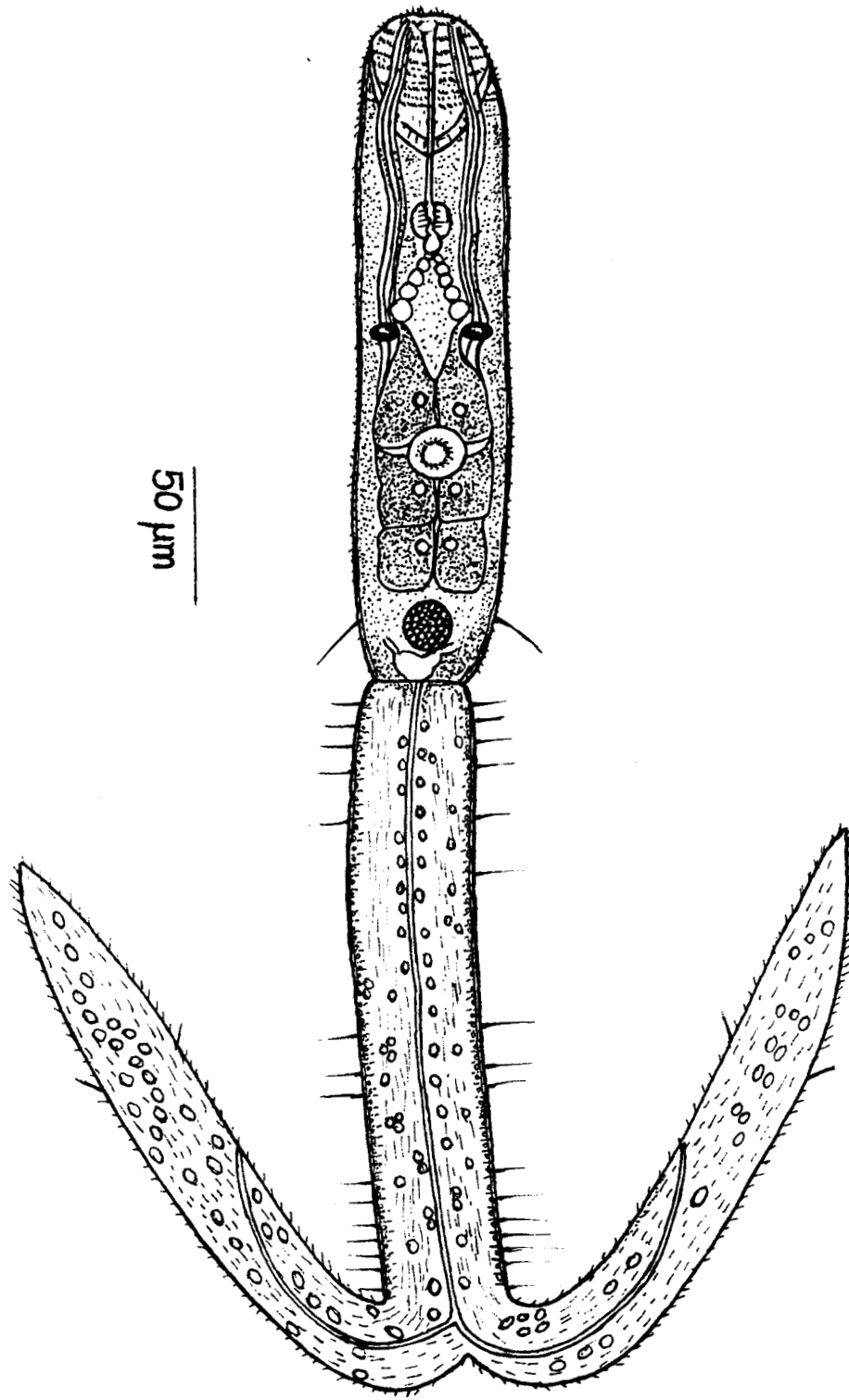


Fig. 10 : Cercaria of *Diplostomum ketupanense* Vidyarthi, 1937

Remarks:

The present cercaria is similar to *Cercariae indicae* II Sewell, 1922 reported from *Thiara tuberculata* in Calcutta. Mohandas (1974a) also recorded this cercaria from the same host in Kerala. Roopa and Janardanan (2001) obtained this cercaria from *Indoplanorbis exustus* collected from Wayanad, Kannur and Kozhikode districts and developed it into adults of *Diplostomum ketupanense*. They established the life cycle of this fluke for the first time showing that *I. exustus* acts as the first intermediate host, the freshwater fish, *Rasbora daniconius* as the second intermediate host and the pond heron, *Ardeola grayii* as the definitive host. The present cercaria undoubtedly belongs to *D. ketupanense*.

The metacercaria of *D. ketupanense* was first reported by Ganapati and Rao (1954) from the muscle tissues of *Catla catla* in Andhra Pradesh, and later by Abraham and Anantaraman (1955) from the same fish in Madras. The present metacercaria is identical with that reported by the above authors and subsequently by Roopa and Janardanan (2001).

Adults of *D. ketupanense* were reported by Vidyarthi (1937) from *Ketupa zeylonensis hardwieki*, Srivastava (1954) from *Ardeola grayii*, Ganapati and Rao (1962) from *A. grayii* and *Bubulcus ibis*, and Roopa and Janardanan (2001) from *A. grayii*. The adults could not be recovered during the present study.

Cercaria sp. V Malabar n. sp.

Infection by this cercaria was found in 3 of 234 *Thiara tuberculata* collected from Nilambur in Malappuram district during April 2001. Snails collected from all other localities were free from this infection.

The cercariae emerged from the snails immediately after sunrise, and emergence continued till evening. It was found actively swimming for a long time with its tail directed forward. During rest, the body and tail stem remained vertically downward with the furcae spreading maximally apart. It also exhibited creeping movements on the bottom of the container.

Description: (Figs. 11-12a, Table 14)

Longifurcate, pharyngeate, distome cercaria. Body elongate-oval, spinose, 198-330 (252) x 46-102 (63) in size; spines prominent, arranged in regular rows; 5 pairs of sensory hairs on body surface. Tail stem 165-210 (183) long, 33-50 (35) wide, aspinose, with 4 pairs of sensory hairs. Caudal bodies absent in tail stem. Furcae almost equal in length to tail stem, spinose, 155-231 (182) x 13-40 (20) in size; 3 pairs of sensory hairs present on lateral margins of each furca. Anterior organ pear-shaped, highly muscular, measured 42-66 (58) x 26-30 (27), with 8 to 9 rows of prominent, backwardly directed spines on their anterior half. Four prominent spines directed anteriorly and arranged parallel to each other present on

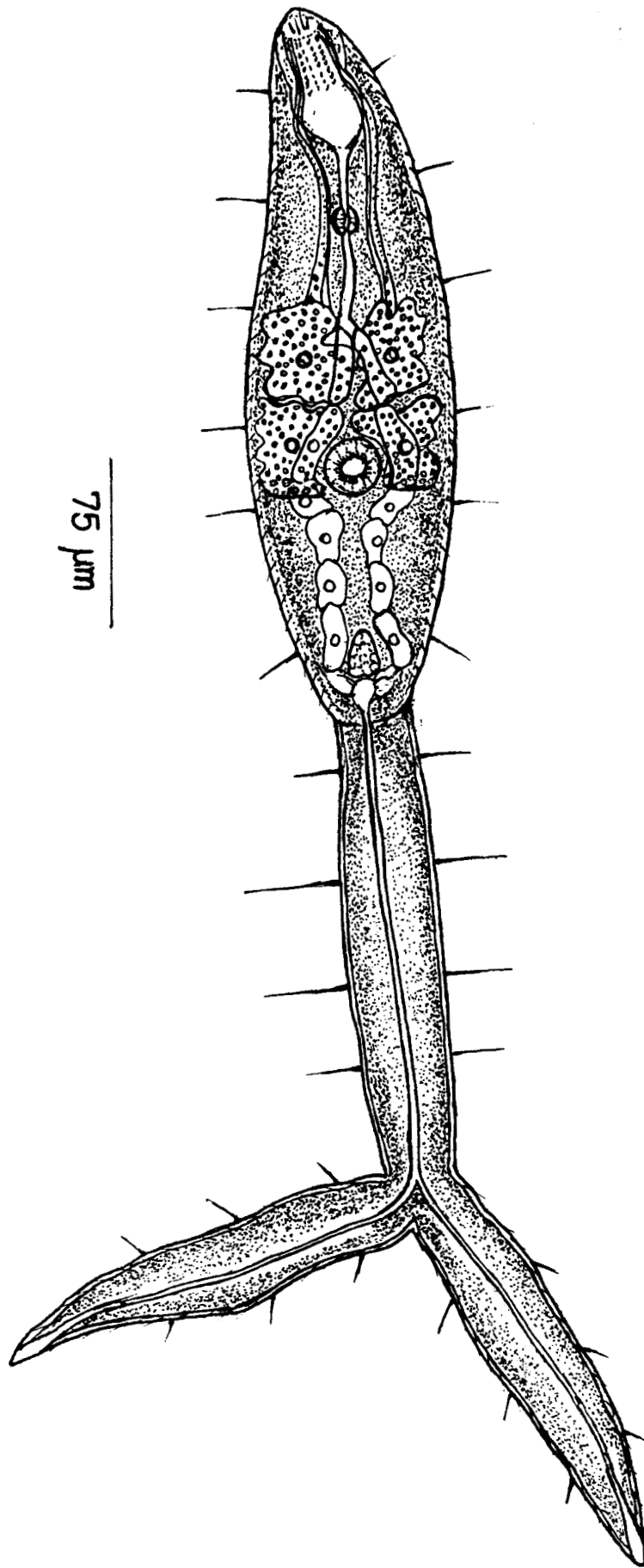
pre-oral lobe. Acetabulum post-equatorial, circular, 19-29 (23) in diameter, with its aperture bordered by a circle of 24 spines.

Mouth subterminal. Prepharynx narrow, long; pharynx muscular, globular, 9-14 (11) in diameter. Oesophagus long. Caeca extend to near posterior end of body; each caecum with 6 cells. Penetration glands 2 pairs, located between caecal bifurcation and posterior margin of acetabulum. Each gland lobed, with coarsely granular contents and large nucleus; gland ducts on each side associate, pass forward, enter laterally into the anterior organ to open at the tip of it. Genital primordium consists of conical mass of cells situated in front of excretory bladder.

Excretory bladder trilobed, at posterior end of body. Symmetrically arranged 8 pairs of flame cells in body and 2 pairs in tail. Flame cell formula, $2[(2+2) + (2+2) + (2)] = 20$. Caudal excretory duct passes through the entire length of tail stem, bifurcates and opens out through pores at the tip of each furca.

Sporocyst: (Fig. 12b)

Sporocysts were found entangled in the hepatopancreas of *Thiara tuberculata*. Body white, long, thread-like, 4368-6428 (5777) long, 140-219 (192) wide. Sporocyst contains numerous fully developed cercariae and a few germ balls at different stages of development.



75 μ m

Fig. 11 : *Cercaria* sp. V Malabar n. sp.

Table 14. Measurements of *Cercaria* sp. V Malabar n. sp.

Characters	Range	Mean
Body	198 - 330 x 46- 102	252 x 63
Tail	165 - 210 x 33 - 50	183 x 35
Furca	155 - 231 x 13 - 40	182 x 20
Anterior organ	42 - 66 x 26 - 30	58 x 27
Acetabulum	19 - 29	23
Pharynx	9 - 14	11
Flame cell formula	$2[(2+2)+(2+2)+(2)] = 20$	

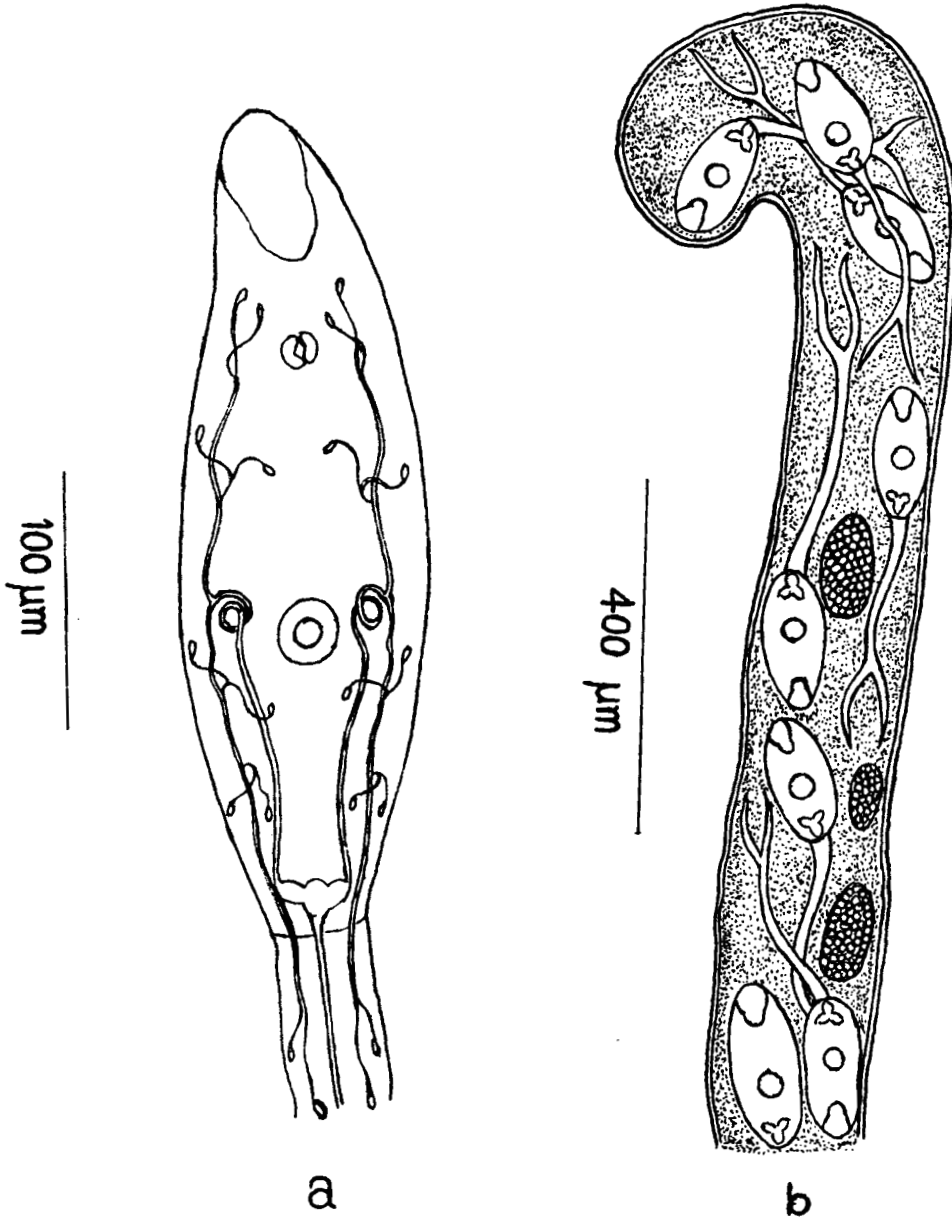


Fig. 12 : *Cercaria* sp. V Malabar n. sp.
a. Excretory system; b. Sporocyst

- Snail host : *Thiara tuberculata* (Mueller)
- Site of infection : Hepatopancreas
- Locality : Nilambur in Malappuram district (Kerala)
- Period of collection : April 2001.
- Prevalence : Three of 1161 (0.26%) snails examined were infected.

Remarks:

The present longifurcate, pharyngeate cercaria is closely comparable to *Cercariae indicae* I Sewell, 1922, *Cercariae indicae* XXII Sewell, 1922, Cercaria of *Cotylurus cornutus* (Rud., 1808) Szidat, 1929 (by Wesenberg-Lund, 1934), Cercaria of *Cotylurus brevis* Dubois *et* Rusch, 1950 (by Dubois, 1934) and cercaria of *Pharyngostomum cordatum* (Diesing, 1850) Ciurea, 1922 (by Wallace, 1939) in having 2 pairs of pre- and/or para-acetabular penetration glands. However, it shows significant differences from the above five species of cercariae in several respects.

The present form could be distinguished from *C. indicae* I and *C. indicae* XXII in the nature and extent of caeca, and in having sensory hairs on tail. *C. indicae* I disagrees further with the present form in the nature of penetration glands, and *C. indicae* XXII in having cephalic glands and caudal bodies. Presence

of non-pigmented eyespots in cercariae of *C. cornuatus* and *C. brevis* makes them distinct from the present form. Cercaria of *P. cordatum* is distinctly different in the number and distribution of sensory hairs on the tail stem and in the number of flame cells. Besides, the cellular caeca and lobed penetration glands found in the present cercaria make it different from the above cercariae.

In the light of these observations the present cercaria is considered new and the name *Cercaria* sp. V Malabar n. sp. is proposed for it.

***Cercaria of Fischoederius elongatus* (Poirier, 1883)**

Stiles and Goldberger, 1910

(Fig. 13)

Host	:	<i>Lymnaea luteola</i> Lamarck
Locality	:	Cherukunnu, Thazhe Chovva, Andalur and Kanhileri in Kannur district; Feroke in Kozhikode district; Anayarangadi, Azhinjilam and Nilambur in Malappuram district; Malampuzha in Palakkad district.
Period of collection	:	November 1997; January to March 1998; December 2000; October – November 2001.
Prevalence	:	Two hundred and seventy-two of 1943 (13.9%) snails examined were infected.

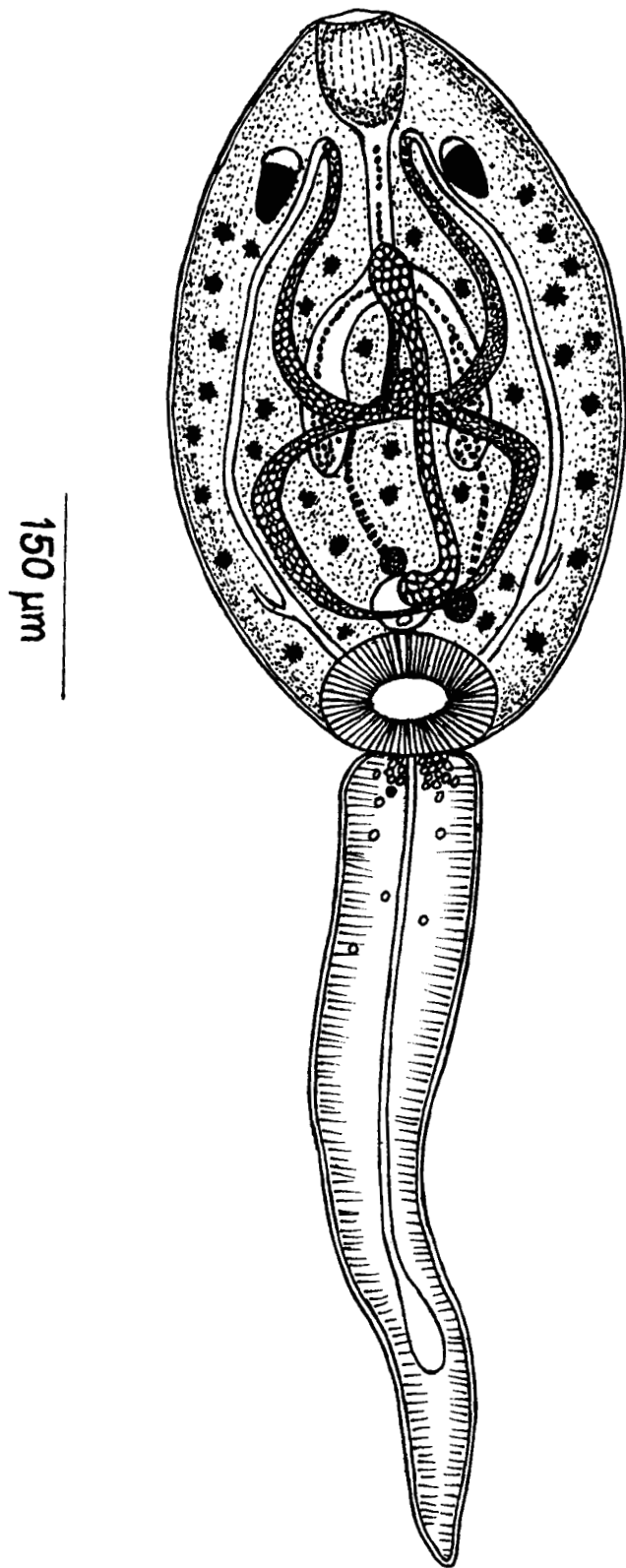


Fig. 13 : Cercaria of *Fiscoederius elongatus* (Poirier, 1883)
Stiles and Goldberger, 1910

Remarks:

The cercaria closely resembled *Cercariae indicae* XXIX Sewell, 1922 in all respects. Sewell (1922) first obtained it from *Lymnaea acuminata* and *Gyraulus euphraticus* in Calcutta and from *L. succinea* in Manantoddy. Rao and Ayyar (1932), and Vaidyanathan (1941) obtained it from *L. luteola* in Madras and Bhalerao (1943) in Hyderabad.

Rao and Ayyar (1932), by feeding experiments, proved that *Cercariae indicae* XXIX Sewell, 1922 is the larval form of *Fischoederius elongatus*. This finding was later confirmed by Vaidyanathan (1941). A detailed account of the life cycle of *F. elongatus* was first presented by Mukherjee (1966a) who implicated *L. luteola* as the snail host and cow and buffalo as the definitive hosts.

Cercaria of *Cotylophoron indicum* Stiles and Goldberger, 1910

(Fig. 14)

Host	:	<i>Indoplanorbis exustus</i> (Deshayes)
Locality	:	Pappinassery, Pallikunnu, Palayad, Peralassery and Manantheri in Kannur district; Feroke and Nallur in Kozhikode district; Azhinjilam and Kizhuparamba in Malappuram district

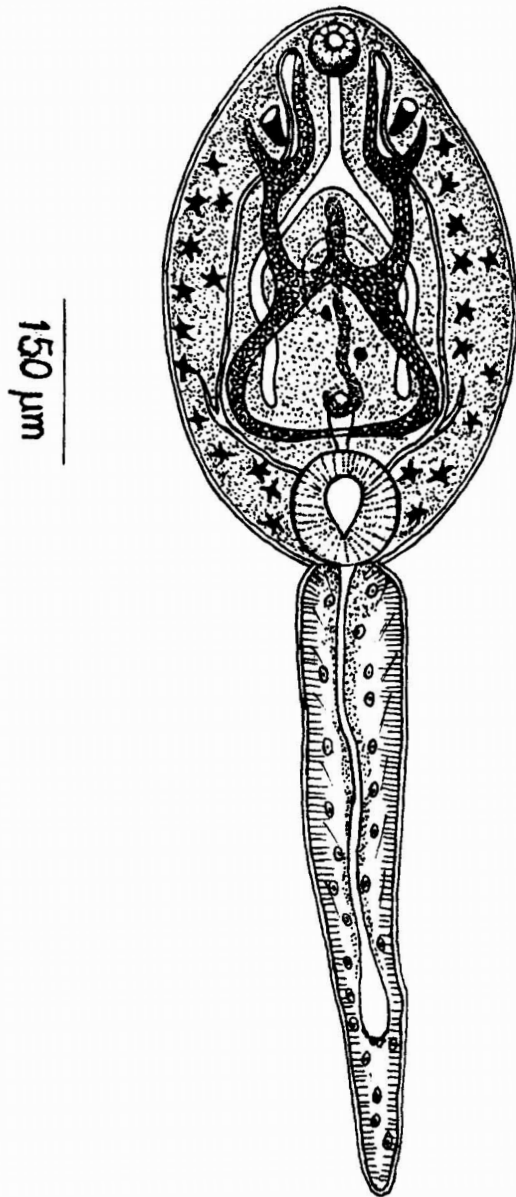


Fig. 14 : Cercaria of *Cotylophoron indicum*
Stiles and Goldberger, 1910

- Period of collection : September – October 1996; December 1998;
November – December 2000; November 2001.
- Prevalence : One hundred and six of 2545 (4.17%) snails examined
were infected.

Remarks:

This amphistome cercaria agreed fully with the description of *Cercariae indicae* XXVI Sewell, 1922 infecting *I. exustus* collected from Wayanad plateau and from the foot of westernghat near Calicut. Subsequently, Rao (1932c) and Anantaraman and Balasubramaniam (1949) obtained the cercaria from the same snail species in Madras city and Bhalerao (1945) from Hyderabad. Experiments conducted by Mukherjee (1960b) proved that *Cercariae indicae* XXVI is the larval form of *Cotylophoron indicum*. Later, complete life cycle of the species was studied by Mukherjee (1968) who showed that the cercariae emerged from *I. exustus*, encysted on grass blades, and then developed into adults in ruminants.

Cercaria of *Echinostoma revolutum* (Frolich, 1802) Dietz, 1909

(Fig. 15)

- Host : *Lymnaea luteola* Lamarck
- Locality : Kanhangad and Theerthamkara in Kasaragod district;

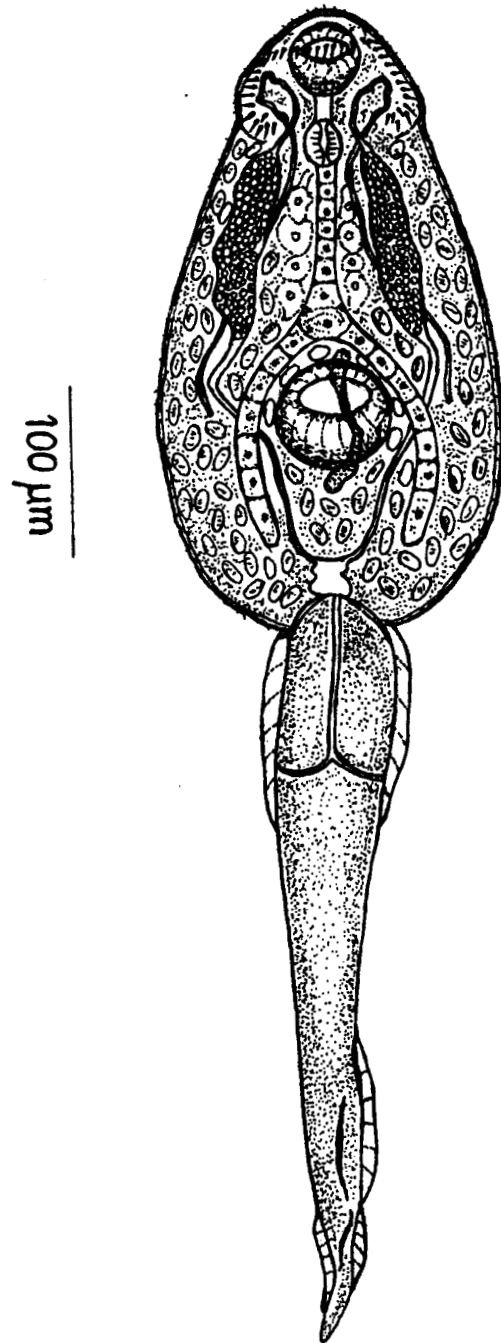


Fig. 15 : Cercaria of *Echinostoma revolutum* (Frölich, 1802)
Dietz, 1909

Patteri in Kannur district; Vadakara in Kozhikode district; Iruvetty in Malappuram district.

Period of collection : November, 1997; December, 2000; November 2001.
Prevalence : Thirty-one of 1943 (1.6%) snails examined were infected.

Natural infections by the metacercariae were found in *L. luteola*, *Indoplanorbis exustus* and *Bellamya dissimilis* collected from the above localities. The cysts were found in the hepatopancreas and gonads of these snails.

Remarks:

The present cercaria is identical in all respects of its structure to that of *Echinostoma revolutum* (Frolich, 1802) Dietz, 1909. Cercariae of this species were reported in India by Peter (1955b) from *Lymnaea luteola f. succinea*, Patnaik and Ray (1966) from *L. auricularia* var. *rufescens* and Mukherjee (1992) from *L. auricularia* var. *rufescens*.

Mohandas (1973a) established the life cycle of a species of *Echinostoma* infecting the domestic fowl, *Gallus gallus* and named it *E. ivaniosi*. This fluke utilized *Lymnaea luteola* and *L. acuminata* as the first intermediate hosts, and *L. luteola*, *L. acuminata* and *I. exustus* as the second intermediate hosts. But based on extensive studies made on adult and larval morphology and biology of *E. revolutum*, Kanev (1994) synonymised *E. ivaniosi* with *E. revolutum*.

A comparison of characters of the present cercaria, and that described for *E. ivaniosi* by Mohandas (1973a) with that of *E. revolutum* presented by Kanev (1994) shows that these cercariae are identical and the decision of Kanev to treat *E. ivaniosi* as a synonym of *E. revolutum* is valid.

Cercaria of *Petasiger variospinosus* (Odhner, 1910)

Yamaguti, 1933

Host	:	<i>Indoplanorbis exustus</i> (Deshayes)
Locality	:	Padanakkad in Kasaragod district; Pallikunnu and Palayad in Kannur district; Payyoli in Kozhikode district; Kizhuparamba in Malappuram district.
Period of collection	:	May 2000 to December 2001.
Prevalence	:	Thirty-six of 2545 (1.41%) snails examined were infected.

Natural infections by the cercariae of *Petasiger variospinosus* (Odhner, 1910) Yamaguti, 1933 were found in *I. exustus* collected from different freshwater bodies at Padanakkad, Pallikunnu, Palayad, Payyoli and Kizhuparamba. The cercariae encysted in freshwater fishes, *Esomus barbatus*, *Rasbora daniconius*, *Puntius parrah*, *Mystus malabaricus* and *Aplocheilus lineatus*, in the same biotopes where the cercarial infections were present. Metacercariae, obtained from natural

infections or those from experimentally infected fishes and fed to the little cormorant, *Phalacrocorax niger*, developed into adults. Natural infections with adult flukes were also obtained from the duodenum and small intestine of *P. niger*. The life cycle of *P. variospinosus* has been successfully established in the laboratory. An article on the life cycle of *P. variospinosus* has been accepted for publication in Rivista di Parassitologia (3/2002, Vol. XIX). A copy of the paper is included under Appendix II.

***Cercaria* sp. VI Malabar n. sp.**

Natural infections with this cercariae were found in the apple snail *Pila virens* collected from paddy fields at Palayad in Kannur district and Kizhuparamba in Malappuram district during November 2000 to January 2001. The prevalence of infection was 1.01% in a total of 889 specimens examined.

Cercarial emergence was most pronounced during morning hours but continued in lesser numbers till evening. Cercariae were found swimming actively with brief periods of rest. At rest, the anterior part of cercarial body bent ventrally giving a tetragonal appearance and the tail remained contracted. Cercariae exhibited crawling movements, at the bottom of the container.

Description: (Figs. 16-17a, Table 15)

Echinostome cercaria. Body elliptical, spinose, measured 198 - 297 (233) x 90-172 (141). Collar and collar spine undifferentiated. Tail aspinose, provided with cuticular annulations, without finfolds, measured 66-160 (99) x 36-50 (43). Oral sucker circular, subterminal, 42-53 (49) in diameter, with a semicircular transverse row of 11 toothlets on its dorsal wall and 9 on its ventral wall. Acetabulum post-equatorial, 39-53(47) in diameter, with a circle of 37-39 scale-like spinelets bordering the aperture.

Mouth subterminal. Prepharynx 6.6-15.2 (9.9) long. Pharynx well developed, 16-20 (18) x 16-20 (17) in size. Oesophagus 26-83 (45) long, bifurcating in front of acetabulum. Caeca 66-126 (86) long, almost touching the antero-lateral margin of excretory bladder.

Penetration glands 5 pairs, along oesophagus; gland ducts open in dorsal lip of oral sucker. Cystogenous glands two types, distributed throughout body except at oral sucker region; one type filled with coarse granular, the other type of fine granules. Genital primordia consist of two masses of cells, one at anterior margin and the other at posterior margin of acetabulum.

Excretory system stenostomate, with bipartite excretory bladder; main collecting ducts distended between acetabular and pharyngeal levels, each containing 10-18 excretory concretions. Flame cell formula, $2[(2+2) + (2+2)] = 16$. The caudal excretory duct extends to middle of tail and opens out laterally.

50 μ m

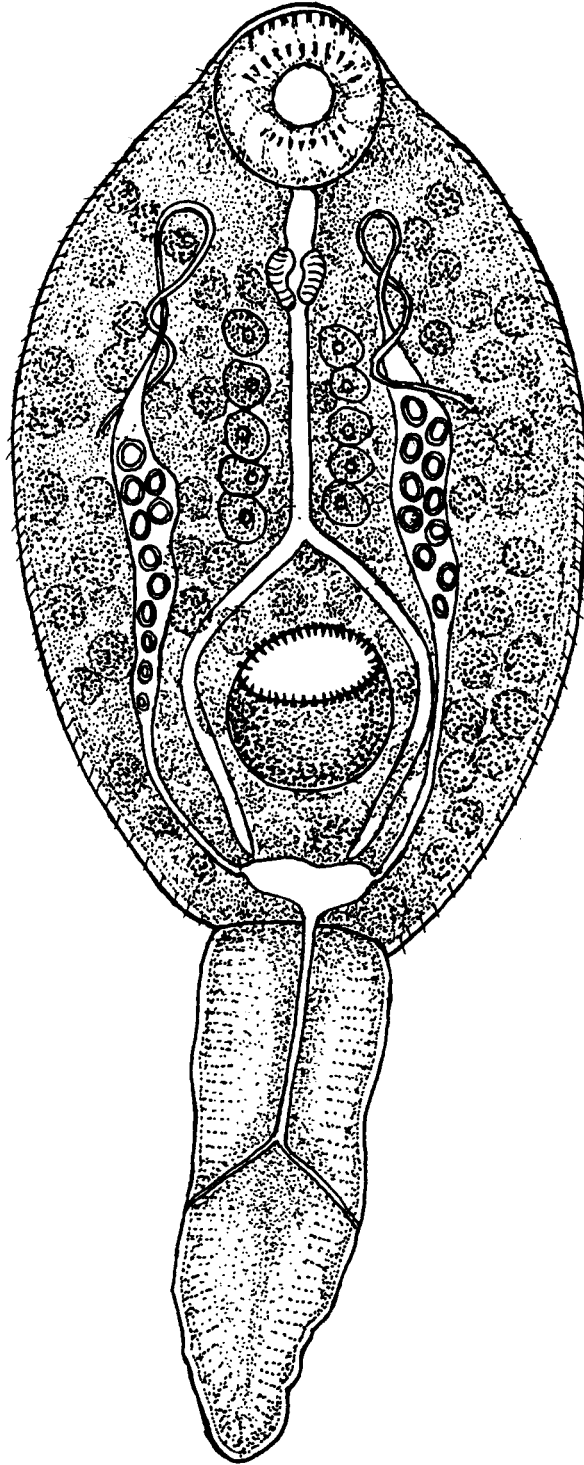


Fig. 16 : *Cercaria* sp. VI Malabar n. sp.

Table 15. Measurements of *Cercaria* sp. VI Malabar n. sp.

Characters	Range	Mean
Body	198 - 297 x 90 - 172	233 x 141
Tail	66 - 160 x 36 - 50	99 x 43
Oral sucker	42 - 53	49
Acetabulum	39 - 53	47
Prepharynx	6.6 - 15.2	9.9
Pharynx	16 - 20 x 16 - 20	18 x 17
Oesophagus	26 - 83	45
Caeca	66 - 126	86
Flame cell formula	$2[(2+2)+(2+2)] = 16$	

Redia: (Fig. 17b)

Rediae were recovered from the hepatopancreas of *Pila virens*. Body sausage-shaped, light yellow, with prominent collar and locomotor organ; measured 462-1248 (822). Pharynx globular, 38-47 (42) in size. Gut 192-578 (314) long, 46-131 (78) wide, filled with yellow-brown granules. Rediae contain 9-26 developing cercariae and many germ balls at different stages of development.

Metacercaria: (Fig. 18a-d)

Infection-free freshwater fishes were exposed to cercariae. Successful experimental infections were established in *Danio aequipinnatus*, *Aplocheilichthys lineatus*, *Labeo rohita* and *Macropodus cupanus*. As many as 26 cysts were obtained from a single *M. cupanus* exposed to cercariae. *P. virens* and other snails of the locality were refractory to this infection. Details of development of metacercariae were followed in *M. cupanus* exposed to cercariae.

In experimentally infected fishes, the cercarial body were observed in gills after 2 to 4 h of exposure. Metacercariae developed thin cyst walls within 8 h of exposure; they were similar to cercarial body except for the absence of penetration and cystogenous glands. In fishes examined 1-day-post infection, the metacercariae developed double-layered, transparent cyst walls, and measured 89-116 (106) x 69-99 (82). On the third day, the cysts became slightly larger in size and measured 118-132 (126) x 66-106 (78). On the 6th day the significant change

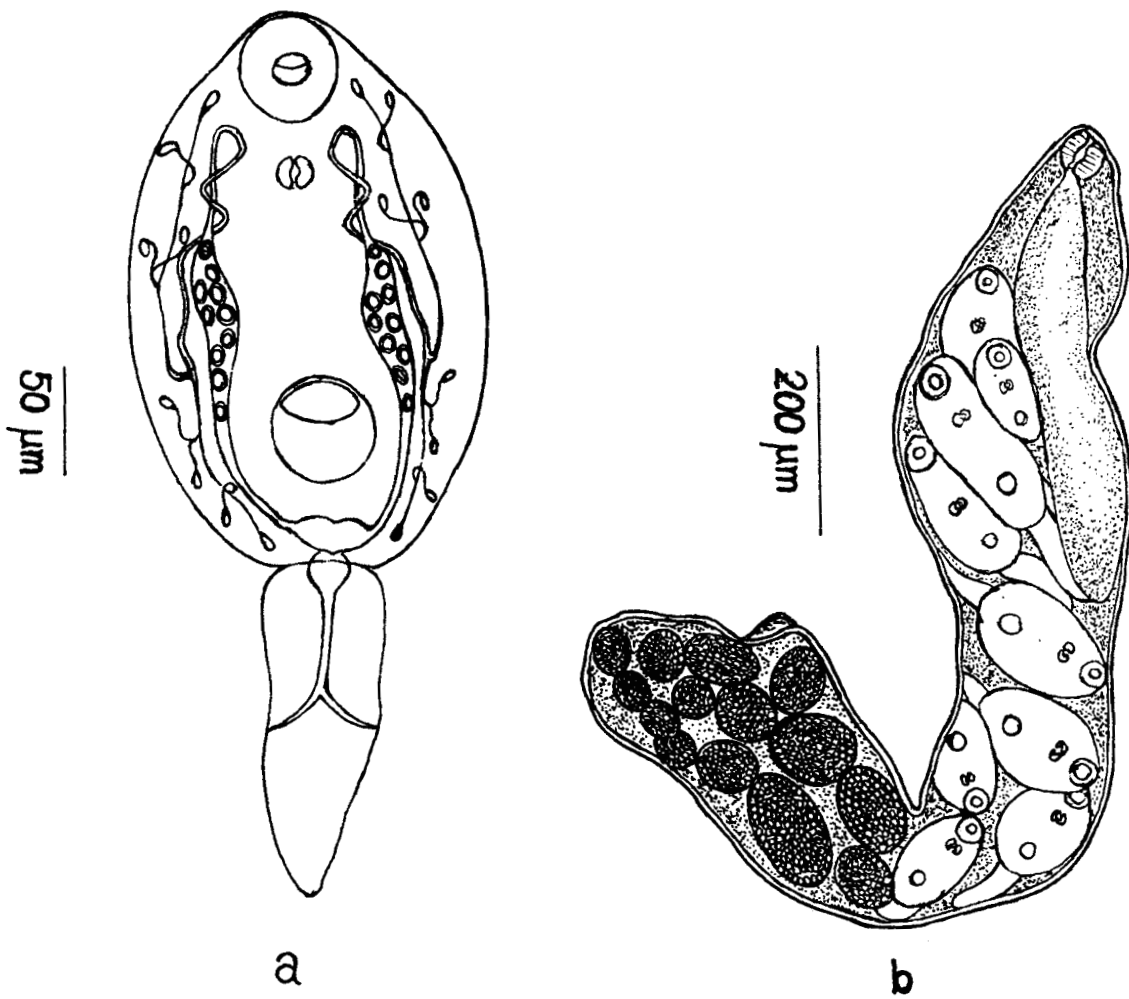


Fig. 17 : *Cercaria* sp. VI Malabar n. sp.
a. Excretory system; b. Redia

noted was the appearance of buds of collar spines; metacercariae measured 120-136 (130) x 89-113 (98). On the 8th day post-infection, the collar spines became prominent; cyst measured 122-136 (132) x 92-114 (100). Developing metacercariae observed on 12th, and 20th day post-infection did not exhibit significant structural changes except a slight increase in size.

On 44th day post-infection, the metacercariae measured 135-150 (140) x 95-116 (106). Excysted larva fusiform, measured 320-350 (339) x 100-108 (102). Collar indistinct, with 24 collar spines arranged in a dorsally interrupted row. The two groups of 4 corner spines measured 6-7.5 (6.7) x 1.5; the 16 dorso-lateral spines measured 6.75-8.25 (7.5) x 1.5 in size. Oral sucker subterminal, 54-58 (56) in diameter, with semicircular transverse rows of 11 toothlets, 3.4-4.5 long, on its dorsal wall and 9 toothlets, 3.3-3.4 long, on its ventral wall. Acetabulum in posterior half of body, 49-52 (49.5) in diameter with a circle of 37-39 spinelets bordering the aperture. Excretory bladder heart shaped, opening terminally. Genital rudiments seen as a mass of cells in post-acetabular region.

Experimental attempts to develop adult flukes by feeding infective metacercariae to one-day-old chicks, white rats, *Ardeola grayii* and *Phalacrocorax niger* were unsuccessful. Attempts to find out the adults in natural hosts were also futile.

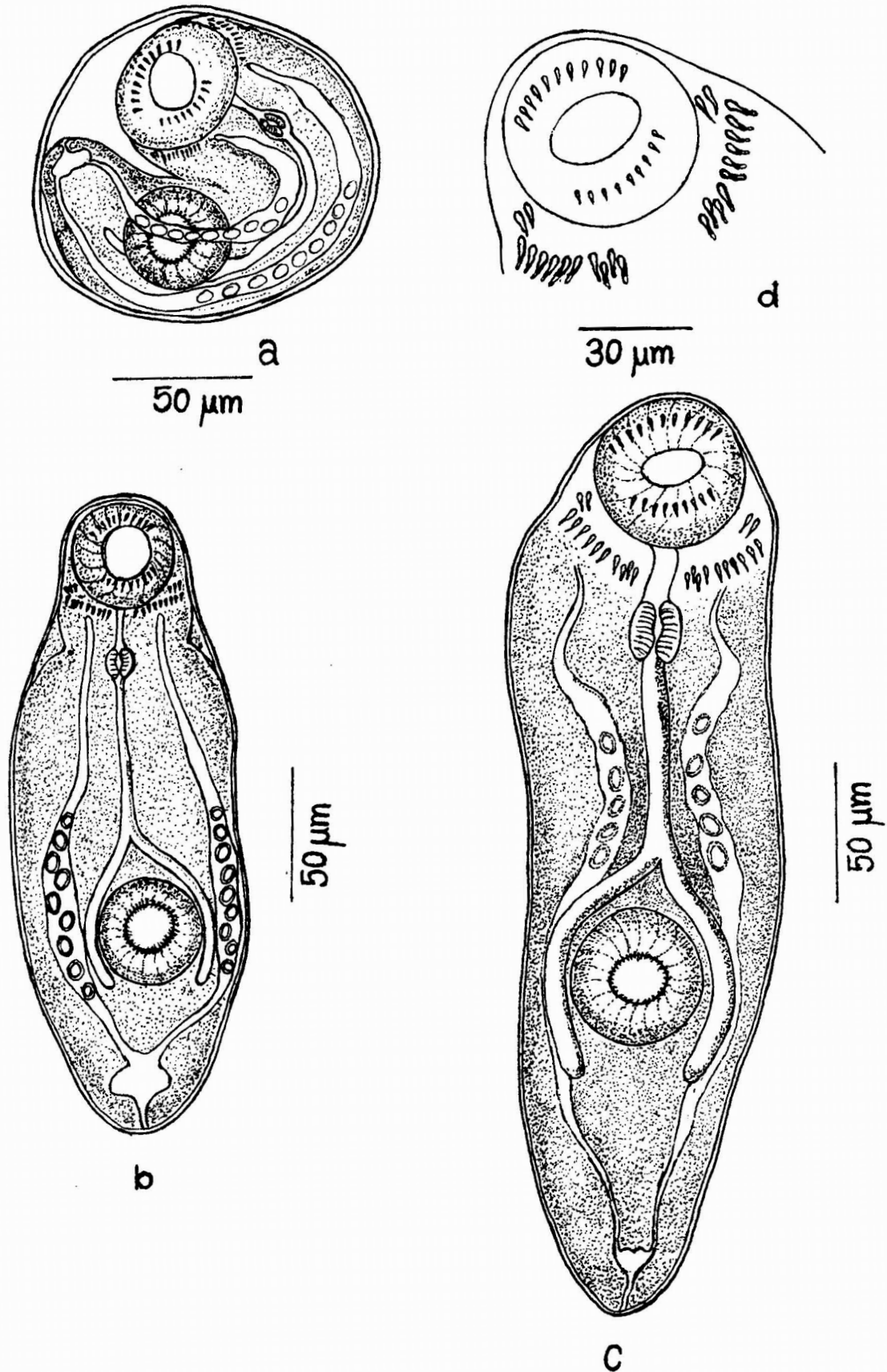


Fig. 18 : Metacercaria of *Cercaria* sp. IV Malabar n. sp.
 a. 6-day-old, encysted; b. 6-day-old, excysted
 c. Collar with collar spines (of 44-day-old metacercaria)
 d. 44-day-old, excysted

- Snail host : *Pila virens* (Lamarck)
- Site of infection : Hepatopancreas
- Locality : Palayad in Kannur district, Kizhuparamba
in Malappuram district (Kerala)
- Period of collection : November 2000 to January 2001.
- Prevalence : Nine of 889 (1.01%) snails examined were
infected.
- Fish hosts : *Danio aequipinnatus* McClelland
Labeo rohita (Hamilton)
Aplocheilus lineatus (Valenciennes)
Macropodus capanus Valenciennes

Remarks:

The present cercaria infecting *Pila virens* belongs to the family Echinostomatidae. Although collar and collar spines were not visible in the cercarial stage, these became prominent in experimentally developed metacercariae. The metacercaria has an indistinct collar with 24 collar spines arranged in dorsally interrupted row. The cercaria is characterised by the presence of a semicircular transverse row of 11 toothlets on the dorsal wall of oral sucker and 9 on the ventral wall. Besides, its acetabulum has a circle of 37-39 scale-like

spinelets bordering the aperture. These type of toothlets and spinelets have not been observed in any of the echinostome cercariae recovered from Indian snails. Cercaria of *Microparyphium kyushuense* reported by Koga (1952) from *Semisulcospira libertina*, *Cercaria redicystica* Tabangui, 1928 from *Pila luzonica* (= *C. leyteensis* no. 25 by Ito, 1977b from *Bellamya philippinensis* and *Pila ampulacea*) and *C. leyteensis* no. 8 Ito, 1977 from *Melanoides tuberculatus* and *Antemelania dactylus* are the other cercariae having toothlets and spinelets on suckers. A comparative study of characters of the three species of cercariae with that of the present form revealed that the cercaria of *M. kyushuense* and *C. redicystica* have aspinose body with sensory papillae, whereas the present form has spinose body without any sensory papillae. Further, the number of toothlets and spinelets on suckers is also different. *C. leyteensis* no.8 is distinct in having a spinose tail which is longer than body, 12 toothlets on dorsal and 14 toothlets on ventral wall of oral sucker and 31 spinelets on acetabulum. There are several other minor characters also which make the 3 cercariae distinct from the form under discussion.

Since the present cercaria cannot be identified with any known echinostome cercariae having toothlets on oral sucker and spinelets on acetabulum, it is considered as new and the name *Cercaria* sp. VI Malabar n. sp. is proposed for it.

The present cercaria developed into metacercaria in the gills of the freshwater fishes *Danio aequipinnatus*, *Labeo rohita*, *Aplocheilus lineatus* and *Macropodus cupanus*. The metacercaria developed 24 dorsally interrupted collar spines on a weakly developed collar. Development of collar spines and retention of toothlets and spinelets on oral sucker and acetabulum respectively at once suggests of its relation to the genus *Microparyphium*. Koga (1952) worked out experimentally the life cycle of *M. kyushuense* with *Semisulcospira libertina* as the first intermediate host, *Misgurnus anguillicaudatus* as the second intermediate host and dog as the definitive host. The metacercariae obtained from the gills of the fish are comparable to the present form, in overall morphology but differ in the number of toothlets and spinelets on suckers and also in the number and arrangement of collar spines.

Experimental attempts to develop adult flukes by feeding infective metacercariae obtained experimentally from fishes to one-day-old chicks, white rats, *Ardeola grayii* and *Phalacrocorax niger* were unsuccessful. Attempts to find out the natural definitive host were also futile. As the metacercariae is related to the genus *Microparyphium* it may be expected to develop into a species belonging *Microparyphium*.

Cercaria sp. VII Malabar n. sp.

Natural infections by the cercariae were found in 13 of 234 (5.56%) *Paludomus transchauricus* collected from irrigation canals at Kanhileri in Kannur district. Infected snails were obtained only during the monsoon period (June to September). Snails collected from all other localities were negative for this infection.

Cercariae emerged mostly during night, and emergence continued in small numbers throughout day-time. Cercariae were positively phototactic, accumulated on the brighter side of the container and exhibited worm-like undulating movements. They remained alive for about 30 h in well water.

Description: (Fig. 19, Table 16)

Distome, macrocercous cercaria. Body oval, white, aspinose, 106-190 (138) long, 61-106 (81) wide. Tail very large, aspinose, transversely striated, measured 561-948 (782) x 76-123 (112) in size; 8 to 12 refractile granules arranged along the central axis of tail. Posterior end of tail modified into an adhesive organ.

Oral sucker terminal, slightly oval, measured 30-46 (36) x 30-37 (32). Acetabulum subspherical near posterior end, measured 36-62 (48) x 34-60 (45). Internal margin of acetabulum bordered by 32 small, scale-like spines.

Mouth terminal. Pre-oral lobe contain 8-10 parallely arranged, tubule like formations. Prepharynx slender, 6-10 (8.8) long. Pharynx muscular, measured 13-32 (26) x 16-31 (23). Oesophagus long, bifurcates at equatorial zone; caeca terminate near posterior end of body. Penetration glands 3 pairs, arranged in a compact manner, around prepharynx. Cystogenous glands numerous, distributed throughout body and filled with rod-shaped contents. Genital primordium spherical, situated in front of acetabulum. Excretory bladder saccular, transversely constricted. Flame cell formula, $2[(2+2)+(2+2)] = 16$. Caudal excretory canal extends up to tip of tail.

Redia: (Fig. 20)

Rediae were recovered from the hepatopancreas of *Paludomus transchauricus*. Body elongate, narrow, light yellow with a prominent collar and a pair of posterior locomotor organs; measured 804-1009 (980) long 160-193 (186) wide. Pharynx globular, 45-62 (52) in diameter, gut extends to the level of locomotor organ, filled with yellow contents. Rediae contain 9-14 cercariae and several germ balls at different stages of development.

Snail host : *Paludomus transchuricus* (Gmelin)
 Site of infection : Hepatopancreas
 Locality : Kanhileri in Kannur district (Kerala)

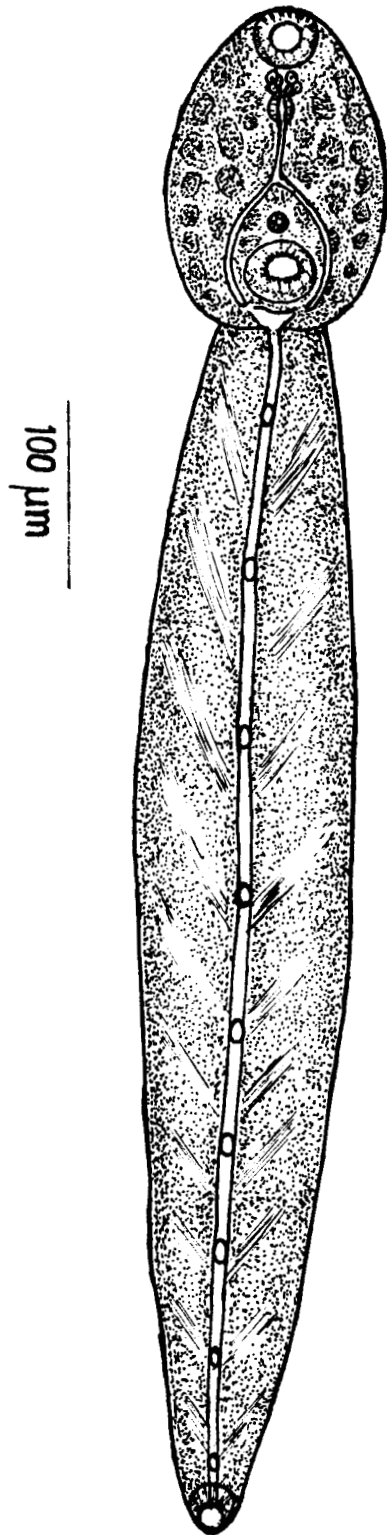


Fig. 19 : *Cercaria* sp. VII Malabar n. sp.

Table 16. Measurements of *Cercaria* sp. VII Malabar n. sp.

Characters	Range	Mean
Body	106 - 190 x 61 - 106	138 x 81
Tail	561 - 948 x 76 - 123	782 x 112
Oral sucker	30 - 46 x 30 - 37	36 x 32
Acetabulum	36 - 62 x 34 - 60	48 x 45
Pharynx	13 - 32 x 16 - 31	26 x 23
Flame cell formula	$2[(2+2)+(2+2)] = 16$	

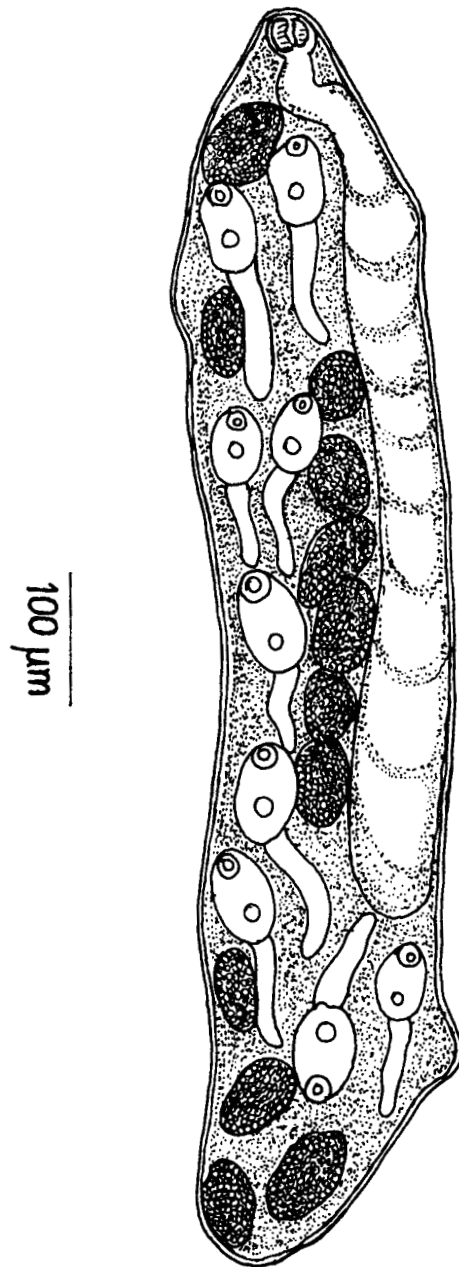


Fig. 20 : *Cercaria* sp. VII Malabar n. sp.
Redia

Period of collection : June to September 1998; June to September 2000.

Prevalence : Thirteen of 1615 (0.80%) snails examined were infected.

Remarks:

The non-ocellate macrocercous cercaria under report needs comparison with the cercaria of *Echinochasmus milavi* Yamaguti, 1939 (by Koga, 1952), *Cercaria chromatocerca* Ito, 1960 and *Cercaria* of *Echinochasmus macrocaudatus* Ditrich, Scholz and Vargas-Vazquez, 1996 (by Ditrich *et al.*, 1996) which have aspinose body, very large and aspinose tail and caeca reaching up to posterior end of body.

Cercaria of *E. milavi* is distinctly different in the number of penetration glands, flame cells and spines on the internal margin of acetabulum and in having tail with wider anterior part and appendix-like posterior part. Presence of sensory hairs on the body and brilliant colouration of the tail make *C. chromatocerca* different from the present form. *Cercaria* of *E. macrocaudatus* has only 2 pairs of penetration glands, whereas the present form has 3 pairs. Further, the opening of its excretory bladder in the first third of tail.

Cercaria komiyani Ito, 1956, *Cercaria nigrocaudata* Ito, 1956 and cercaria of *Galactosomum ussuriense* Oshmarin, 1963 (by Rekharani and Madhavi, 1985b) are the other cercariae showing superficial resemblance to the present form because

of the presence of broad and long tail. But unlike the present form they have sparse body, eye spots and different number of penetration glands. *C. komiyani* disagrees further with the present form in the number of flame cells and in the absence of adhesive organ on tail tip, and *C. nigrocaudata* and cercaria of *G. ussuriense* in the absence of acetabulum.

Considering the differences with the related form, the present form is regarded as new and designated *Cercaria* sp. VII Malabar n. sp.

***Cercaria* sp. VIII Malabar n. sp.**

Natural infections with the cercariae were found in the thiarid snail, *Thiara tuberculata* collected from Nilambur in Malappuram district. Snails collected from all other localities were negative for this infection. Infection was restricted to the period extending from April to June. Of the 1161 snails examined 8 were found to be infected, the prevalence of infection being 0.76%.

Cercariae were found emerging from snails in moderate numbers during morning hours, but emergence in lesser numbers continued throughout day time. They accumulated in large numbers in the surface layer of water. The cercariae were active swimmers; they performed leech-like movements on a glass slide with a thin film of water or at the bottom of the container.

Description: (Figs. 21-22a, Table 17)

Gymnocephalous, distome, non-ocellate cercaria. Body large, elongated, spinose, heavily pigmented, 231-604 (412) long, 135-212 (169) wide. Tail shorter than body, aspinose, attached subventrally, measured 198-363 (246) long, 33-60 (43) wide. Tip of the tail invaginated into a flask-shaped structure with 2 unicellular glands on each side, positioned one above the other.

Oral sucker terminal, round, 28-70 (43) in diameter. Acetabulum pre-equatorial, 33-73 (50) in diameter.

Mouth subterminal; prepharynx absent. Pharynx well-developed, muscular, 16-37(15) in diameter. Oesophagus short, 20-50 (29) in length, bifurcates in front of acetabulum; caeca extend to near posterior end of body. Penetration glands 11 pairs, located on either side of oesophagus; outer two groups composed of 8 glands each and inner groups 3 glands each. Each gland pyriform with round nucleus and granular contents. Ducts from each group associate, pass forward and open out separately at antero-lateral region of mouth. Cystogenous glands numerous, scattered throughout posterior two-third of body, composed of 2 types; one type with coarsely granular contents, arranged in peripheral region of body and the other type with finely granular contents, arranged medially.

Genital primordia consist of two masses of round cells, one at anterior margin and the other at posterior margin of acetabulum. Excretory bladder

50 μ m

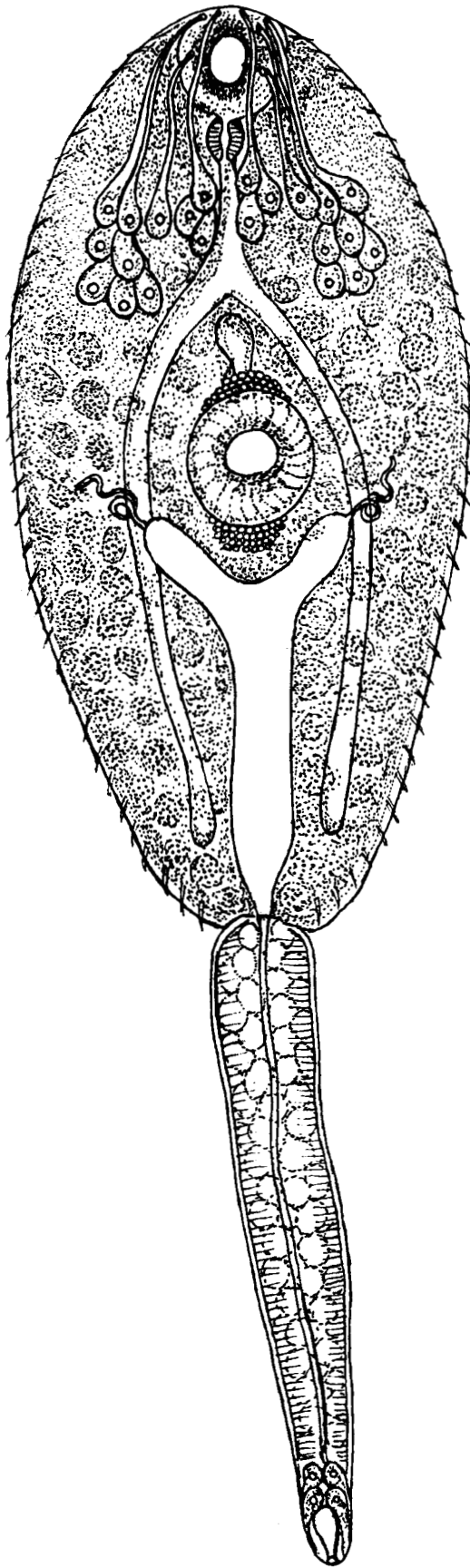


Fig. 21 : *Cercaria* sp. VIII Malabar n. sp.

Table 17. Measurements of *Cercaria* sp. VIII Malabar n. sp.

Characters	Range	Mean
Body	231 - 604 x 135 - 212	412 x 169
Tail	198 - 363 x 33 - 60	246 x 43
Oral sucker	28 - 70	43
Acetabulum	33 - 73	50
Pharynx	16 - 37	15
Oesophagus	20 - 50	29
Flame cell formula	$2[(2+2)+(2+2+2)] = 20$	

'Y' shaped, extending from posterior end of body to post-acetabular level. The main excretory duct arises from each diverticulum, takes a convoluted path up to equatorial region and bifurcates into anterior and posterior collecting ducts. Flame cell formula, $2[(2+2) + (2+2+ 2)] = 20$. Caudal excretory duct runs through tail and opens out at its tip.

Sporocyst: (Fig. 22b)

Body large, sausage-shaped, 936-1888 (1327) long, 191-406 (298) wide. Sporocysts contain 5-13 developing cercariae and several germ balls at different stages of development.

Snail host : *Thiara tuberculata* (Mueller)

Site of infection : Hepatopancreas

Locality : Nilambur in Malappuram district (Kerala)

Period of collection : April to June 2001.

Prevalence : Eight of 1161 (0.76%) snails examined were infected.

Remarks:

Of the various gymnocephalous cercariae with 'Y'-shaped excretory bladder, spinose body and aspinose tail, the present form comes close to

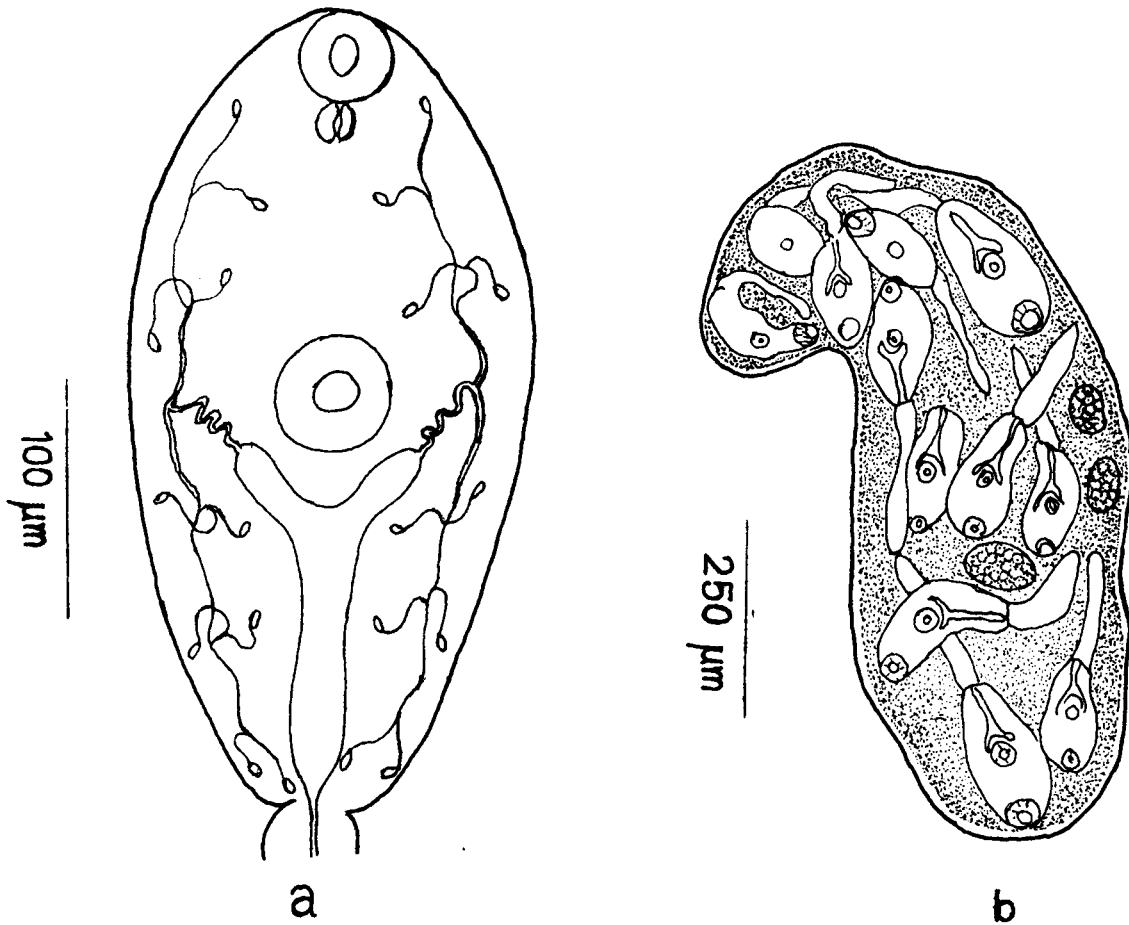


Fig. 22 : *Cercaria* sp. VIII Malabar n. sp.
a. Excretory system; b. Sporocyst

Cecariae indicae XIV Sewell, 1922, *Cercaria tuniforka* Premvati, 1954, *Cercaria Lavantina* 9 Gold and Lengy, 1974 and *Cercaria leyteensis* no. 10 Ito, 1977 in overall morphology and in the snail host used. But the flask-shaped invagination and unicellular glands present at the tail tip make the present cercaria distinct from the others. Further, in the number and arrangement of penetration glands and in the extent of caeca, the present form is different from *C. tuniforka*, *C. Lavantina* 9 and *C. leyteensis* no.10.

Cercariae indicae IV Sewell, 1922, *Cercaria ratnagiriensis* Peter, 1954, *Cercaria caudiglandula* Premvati, 1954, cercaria of *Philophthalmus gralli* Mathis and Leger, 1910 (by Alicata and Noda, 1960; Ching, 1961; Ismail and Arif, 1992) and *Cercaria* sp. 11 Kerala Mohandas, 1979 are the other cercariae characterised by the presence of flask-shaped invagination and unicellular glands at their tail tips. These cercariae, like the present form, have spinose body, aspinose tail and penetration glands located on either side of oesophagus, and therefore, need comparison with it. But the large 'Y'-shaped excretory bladder, extending from posterior end of body to post acetabulum level, of the present forms makes it distinct from the others.

As the present cercaria cannot be identified with any other related cercaria reported earlier, it is considered as new and the name *Cercaria* sp. VIII Malabar n.sp. is proposed for it.

***Cercaria* sp. IX Malabar n. sp.**

Infection by this cercaria was found in the snail, *Paludomus transchauricus* collected from irrigation canals and rivulets at Kanhileri in Kannur district of Kerala during October-November 2000. Infection was rare and only 6 of 1615 snails examined were infected, the prevalence of infection being 0.37%.

The cercariae emerged in small numbers throughout day and night, but in large numbers during afternoon hours. Cercariae swam actively by wriggling movements of body and tail. At rest they remained suspended in the water column. Cercariae exhibited creeping movements at the bottom of the container.

Description: (Figs. 23-24a, Table 18)

Virgulate xiphidiocercaria. Body elongate-oval, spinose, measured 92-149 (122) long, 56-85 (72) wide. Tail spinose, spines larger than body spines; attached postero-ventrally, measured 56-93 (75) long, 13-24 (18) wide. Oral sucker subterminal, large, round, 29-37 (33) in diameter. Stylet prominent, inserted into the roof of oral sucker, 13-14 (13.2) long, 3-3.5 (3.3) wide, stylet walls thick, shoulders moderately developed; virgula organ located at the posterior half of oral sucker; large, bilobed, fused medially, 13-24 (17) long and 13-24 (18) wide. Acetabulum post-equatorial, smaller than oral sucker, 16-24 (18) in diameter.

Mouth subterminal, ventral. Pharynx well-developed, muscular. Oesophagus short, weakly developed; caeca not traceable.

Penetration glands 4 pairs; anterior pair pre-acetabular, second pair para-acetabular and posterior 2 pair post-acetabular; contents of anterior 2 pairs coarsely granular and posterior 2 pairs finely granular. Ducts of penetration glands open individually near stylet shoulders. Genital primordium consists of a mass of round cells located near posterior margin of acetabulum.

Excretory bladder large, 'I'-shaped, with thick wall lined by cells; excretory pore located dorsally at body tail junction. Flame cell formula, $2[(2+2+2) + (2+2+2)] = 24$.

Sporocyst: (Fig. 24b)

Sporocysts were recovered from the hepatopancreas of *Paludomus transchauricus*. Sporocysts colourless, oval to spherical, saccular, 72-215 (168) long, 69-175 (128) wide, containing 7-14 developing cercariae and a few germ balls at various stages of development.

Snail host	:	<i>Paludomus transchauricus</i> (Gmelin)
Site of infection	:	Hepatopancreas
Locality	:	Kanhileri in Kannur district (Kerala)

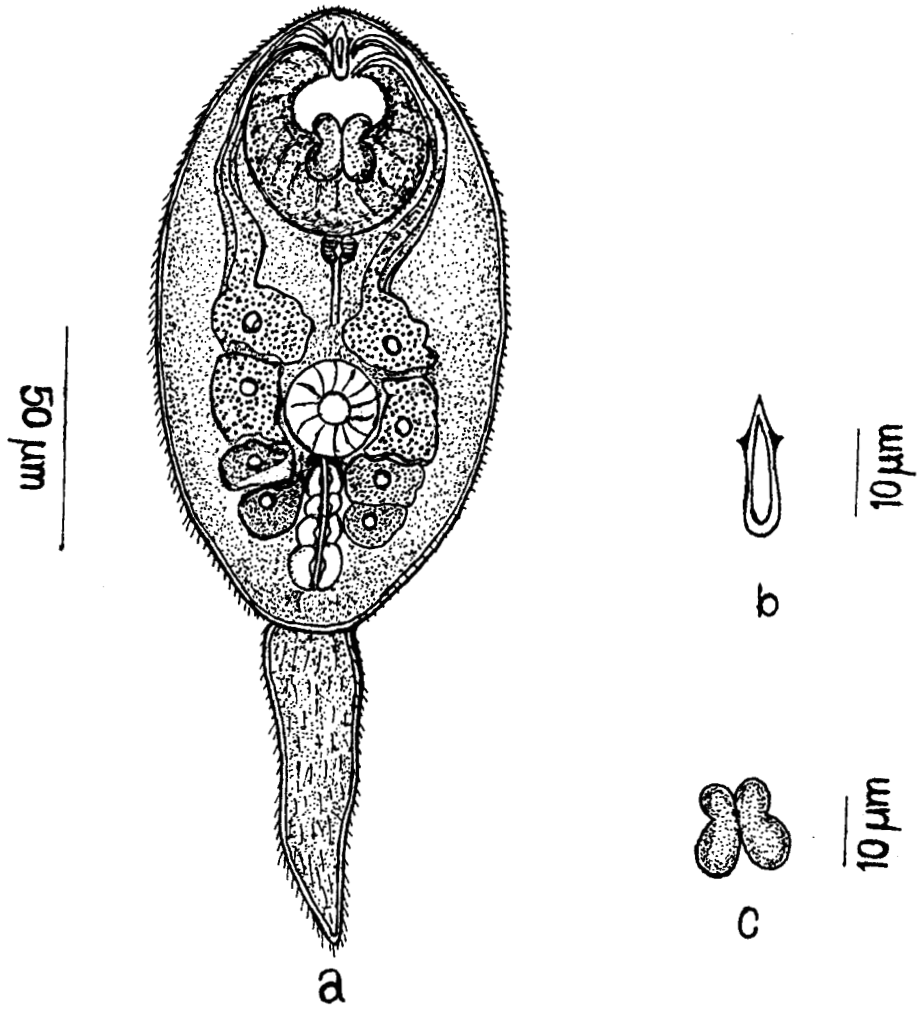


Fig. 23 : *Cercaria* sp. IX Malabar n. sp.
a. Cercaria; b. Stylet; c. Virgula

Table 18. Measurements of *Cercaria* sp. IX Malabar n. sp.

Characters	Range	Mean
Body	92 – 149 x 56 – 85	122 x 72
Tail	56 – 93 x 13 – 24	75 x 18
Oral sucker	29 – 37	33
Stylet	13 – 14 x 3 – 3.5	13.2 x 3.3
Virgula	13 – 24 x 13 – 24	17 x 18
Acetabulum	16 – 24	18
Flame cell formula	$2[(2+2+2)+(2+2+2)] = 24$	

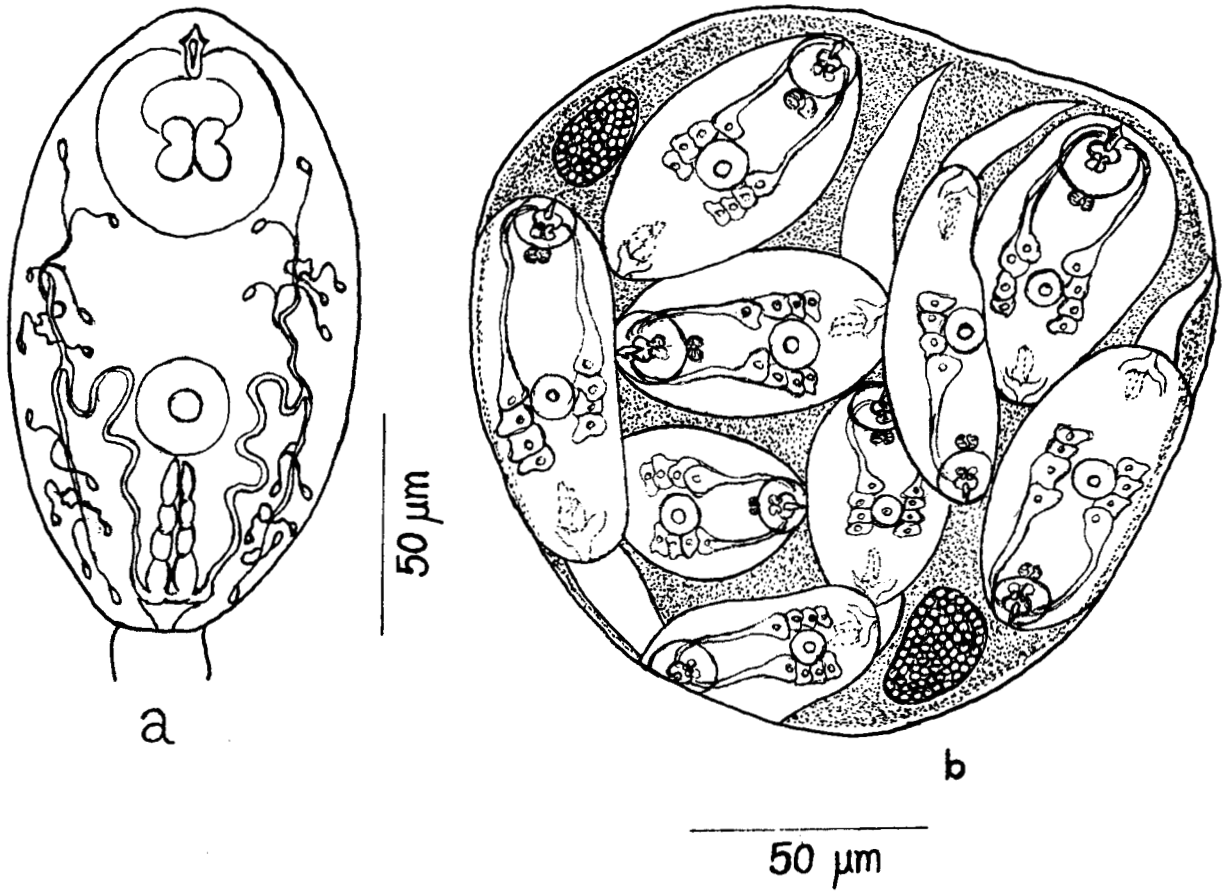


Fig. 24 : *Cercaria* sp. IX Malabar n. sp.
a. Excretory system; b. Sporocyst

Period of collection : October - November 2000.

Prevalence : Six of 1615 (0.37%) snails examined were infected.

Remarks:

Of the known virgulate xiphidiocercariae with spinose body and four pairs of penetration glands, the present form comes close to cercaria of *Allossogonoporus vespertilionis* Macy, 1940 (by Burns, 1961), Cercaria of *Pleurogenoides orientalis* (Srivastava, 1934) (by Murty, 1976; Madhavi *et al.*, 1987), *Cercariae indicae* LXIX Murty, 1976, *Cercaria leyteensis* no. 4 Ito *et al.*, 1977, *Cercaria* sp. XII Kerala Mohandas, 1977, *Cercaria microvirgula* Haseeb and Khan, 1987, Cercaria of *Mehraorchis ranarum* Srivastava, 1934 (by Ratnakumari *et al.*, 1991) and Cercaria of *Pleurogenoides ovatus* Rao, 1977 (by Janardanan and Prasad, 1991).

Cercaria of *A. vespertilionis* differs from the present form in the shape of stylet and virgula organ, nature of penetration gland contents and in having spinose tail. Cercariae of *P. orientalis*, *M. ranarum* and *P. ovatus*, *C. indicae* LXIX, *C. leyteensis* no.4, *C. sp.* XII Kerala and *C. microvirgula* have oval or 'V'-shaped excretory bladder, whereas that of the present form is 'T'-shaped. Cercaria of *P. orientalis*, *C. leyteensis* no. 4, *C. sp.* XII Kerala and *C. microvirgula* further differ from the present cercaria in the nature of penetration gland contents. In the shape of stylet and virgula organ also *C. indicae* LXIX and cercariae of *M.*

ranarum and *P. ovatus* differ from the present cercaria. Besides, the snail host of the present form is different from that of above cercariae.

Hence it appears reasonable to consider the present cercaria as new and the name *Cercaria* sp. IX Malabar n.sp. is proposed for it.

***Cercaria* sp. X Malabar n. sp.**

Infections by this cercaria were found in 8 of 234 (3.4%) thiarid snails, *Paludomus transchauricus*, collected from Kanhileri in Kannur district. The infected snails were collected during November-December 2000. Snails collected from other localities were negative for this cercaria.

The cercariae emerged throughout the day, but peak emergence was noticed during afternoon hours. Newly emerged cercariae swam for a short period by vigorous lashing activity of tail, then sank to the bottom of the container and encysted there.

Description: (Figs. 25-26a, Table 19)

Non-irrigulate xiphidiocercaria. Body elongate-oval, spinose, 85-170 (109) long, 49-66 (57) wide. Tail shorter than body, spinose, highly contractile, attached postero-ventrally; 59-80 (72) long, 9-17 (13) wide. Oral sucker round, 19-33 (27) in diameter. Stylet prominent, inserted into the roof of oral sucker, spear-shaped with fine tip, round base and prominent shoulders at its anterior one third; 18-20

(19.6) long, and 3.3-4.1 (3.5) thick at shoulder region. Acetabulum rudimentary, located in posterior half of body.

Mouth ventral, subterminal. Pharynx well-developed, muscular, 6-10 (8) in diameter.

Penetration glands 3 pairs; anterior 2 pairs pre-acetabular and posterior pair para-acetabular with their ducts opening near stylet shoulders. Glands round to oval with fairly large nuclei. Contents of anterior 2 pairs coarsely granular and posterior pair finely granular. Cystogenous glands numerous, filled with refringent granules, distributed all over the body. Genital primordium represented by a round mass of cells situated anterior to acetabular rudiment.

Excretory bladder 'Y'-shaped with a round base and two broad cornuae; each cornua gives rise to an ascending main excretory duct which takes a convoluted course upto the level of acetabulum and divides into anterior and posterior collecting tubules. Flame cell formula, $2[(2+2) + (2+2)] = 16$. Excretory pore situated at body-tail junction. Caudal excretory canal extend to the tip of tail.

Sporocyst: (Fig. 26b)

Sporocysts were recovered from the hepatopancreas of *Paludomus transchauricus*. Sporocysts oval to elongate, sac-like, 346-577 (473) long, 154-285 (205) wide; enclose 6 to 17 developing cercariae and a few germ balls at various stages of development.

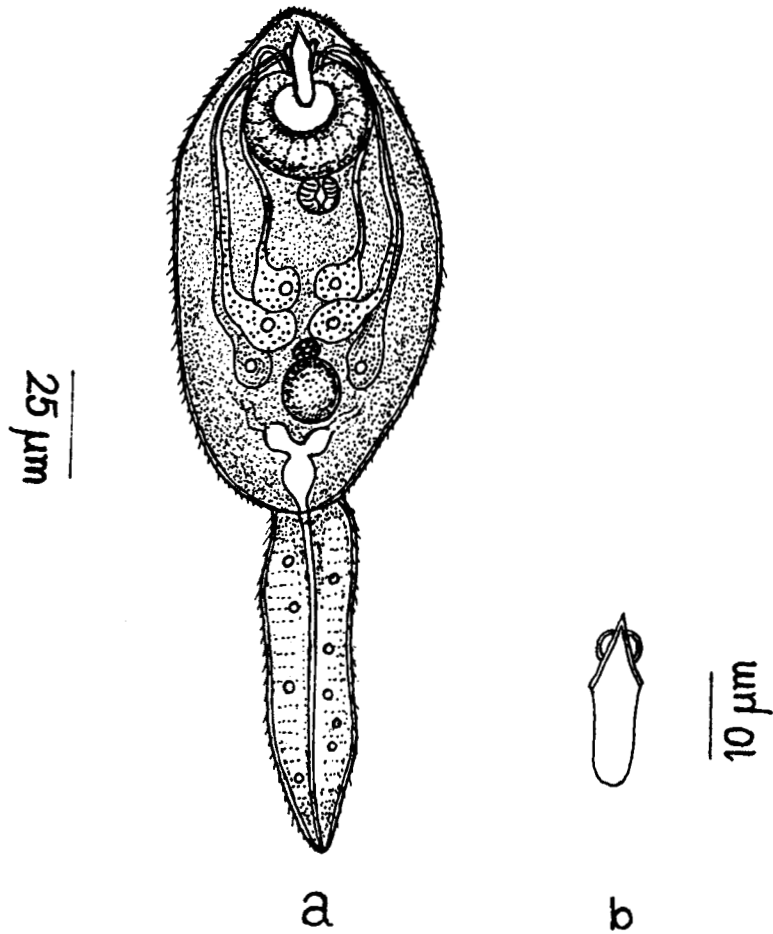


Fig. 25 : *Cercaria* sp. X Malabar n. sp.
a. Cercaria; b. Stylet

Table 19. Measurements of *Cercaria* sp. X Malabar n. sp.

Characters	Range	Mean
Body	85 – 170 x 49 – 66	109 x 57
Tail	59 – 80 x 9 – 17	72 x 13
Oral sucker	19 – 33	27
Stylet	18-20 X 3.3-4.1	19.6 X 3.5
Pharynx	6 – 10	8
Flame cell formula	2 [(2+2)+(2+2)] = 16	

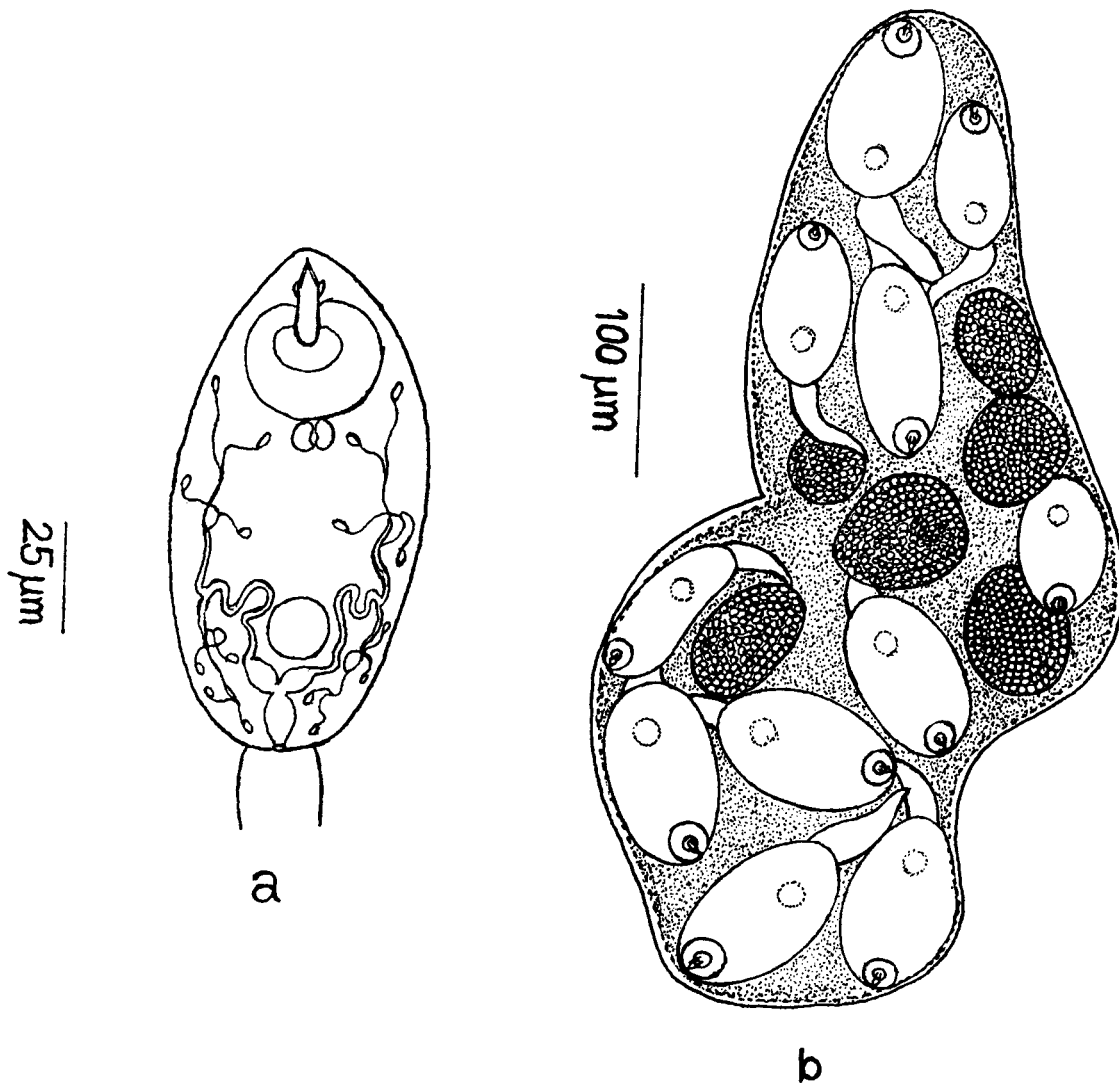


Fig. 26 : *Cercaria* sp. X Malabar n. sp.
a. Excretory system; b. Sporocyst

Snail host	:	<i>Paludomus transchauricus</i> (Gmelin)
Site of infection	:	Hepatopancreas
Locality	:	Kanhileri in Kannur district (Kerala)
Period of collection	:	November - December 2000
Prevalence	:	Eight of 1615 (0.49%) snails examined were infected.

Remarks:

Distome non-virgulate xiphidiocercariae with 3 pairs of medially placed penetration glands and poorly developed acetabulum showing close resemblance to the present cercaria are *Cercariae indicae* LII Sewell, 1922, *Cercaria leyteensis* no.5 Ito *et al.*, 1977, *C. leyteensis* no. 27 Ito, 1977 and *C. visakhapatnamensis* 5 Dhanumkumari *et al.*, 1990. But the present form differs from all these cercariae in one or more of the following characters: body size, stylet shape and size, position and shape of penetration glands and the nature of their contents, number of flame cells, shape of excretory bladder and presence or absence of spines on tail. *C. indicae* LII is distinctly different from the present cercaria in shape and size of stylet, arrangement of penetration glands and nature of their contents, shape of excretory bladder and in having a spinose tail. *C. leyteensis* no.5 has stylet with indistinct shoulders, well developed oesophagus, 4 pairs of sensory hairs on the body and 'U' shaped excretory bladder. These characters make *C. leyteensis* no.5 different from the present form. *C. leyteensis* no. 27 and *C. visakhapatnamensis* 5

also differ from the present cercaria in having aspinose tails, and differential secretions in penetration glands i.e., fine granular secretion in anterior 2 pairs of glands and coarse granular secretion in posterior pair. Further, the snail hosts of the above mentioned cercariae are also different.

As the present cercaria cannot be identified with any other known cercaria, it is considered as new and the name *Cercaria* sp. X Malabar n. sp. is proposed for it.

***Cercaria* sp. XI Malabar n. sp.**

This cercaria was encountered in the thiarid snails, *Thiara tuberculata* collected from Chaliyar in Malappuram district of Kerala. Infection was rare and only 5 out of 1161 snails examined were found to be infected.

Cercariae emerged from snails during morning hours and emergence continued throughout day-time. It swam actively by lashing movements of tail. Prolonged swimming activity was interrupted by brief periods of rest. The cercariae performed creeping movements on a glass slide with a thin film of water or at the bottom of the container.

Description: (Figs. 27-28a, Table 20)

Non-irrigulate xiphidiocercaria. Body oval, spinose, 89-198 (138) long, 66-99 (83) wide. Tail aspinose, highly contractile, attached postero-ventrally;

measured 69-156 (96) long and 13-20 (16) wide. Oral sucker subterminal, large, round, 26-37 (32) in diameter. Stylet inserted into the roof of oral sucker, with pointed tip, prominent shoulders and broadly round base; measured 15.6-18.2 (16.6) long, and 3-4 (3.3) wide at base. Acetabulum post-equatorial, smaller than oral sucker, 16-24 (19) in diameter.

Mouth subterminal, ventral; prepharynx absent. Pharynx globular, muscular, 6.6-9.9 (8.4) in diameter. Oesophagus short; caeca not observed.

Penetration glands 3 pairs in acetabular region; anterior one pair with coarsely granular and posterior 2 pairs with finely granular contents. Each gland pyriform with irregular margin and prominent nucleus. Ducts of the penetration glands open out through pores near the base of stylet. Genital primordia consist of 3 round masses of cells located near posterior margin of acetabulum.

Excretory bladder, thin walled, bicornuate; excretory pore situated dorsally at body tail junction. Flame cell formula, $2[(3+3)+(3+3)] = 24$.

Sporocyst: (Fig. 28b)

Sporocysts were found in the hepatopancreas of *Thiara tuberculata*. Body oval to sausage-shaped, sac-like, 99-231 (161) long, 69-193 (91) wide. Each sporocyst encloses 3-4 fully developed cercariae and 2-5 germ balls at various stages of development.

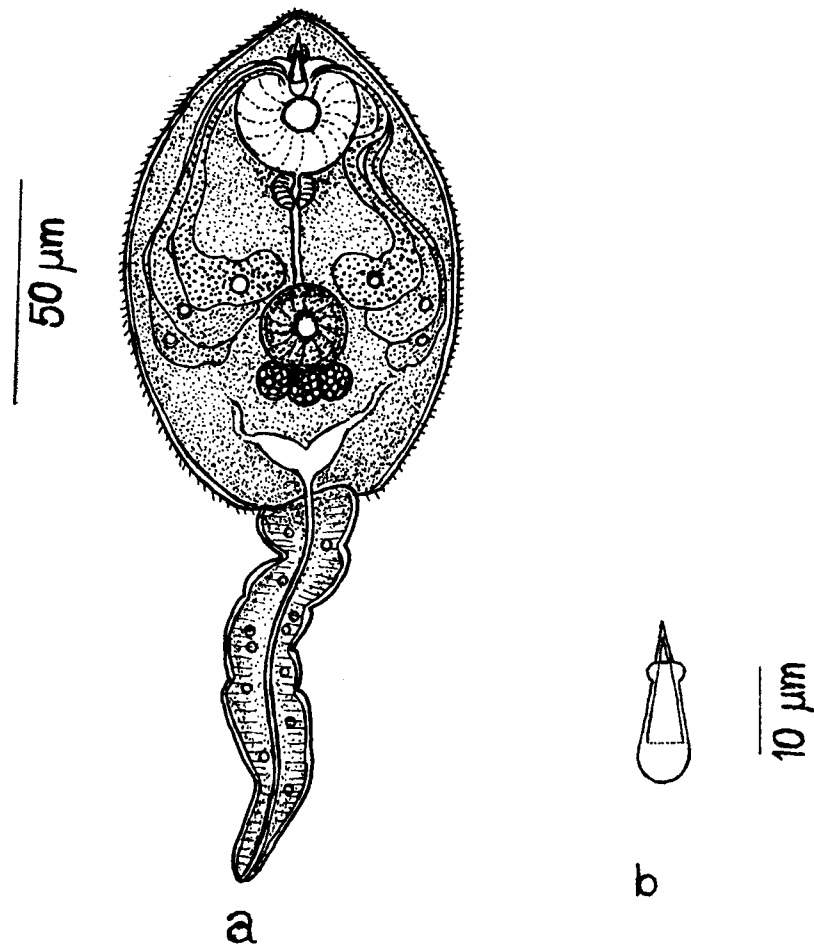


Fig. 27 : *Cercaria* sp. XI Malabar n. sp.
a. Cercaria; b. Stylet

Table 20. Measurements of *Cercaria* sp. XI Malabar n. sp.

Characters	Range	Mean
Body	89 – 198 x 66 – 99	138 x 83
Tail	69 – 156 x 13 – 20	96 x 16
Oral sucker	26 – 37	32
Stylet	15.6 – 18.2 x 3 – 4	16.6 x 3.3
Acetabulum	16 – 24	19
Pharynx	6.6 – 9.9	8.4
Flame cell formula	$2[(3+3)+(3+3)] = 24$	

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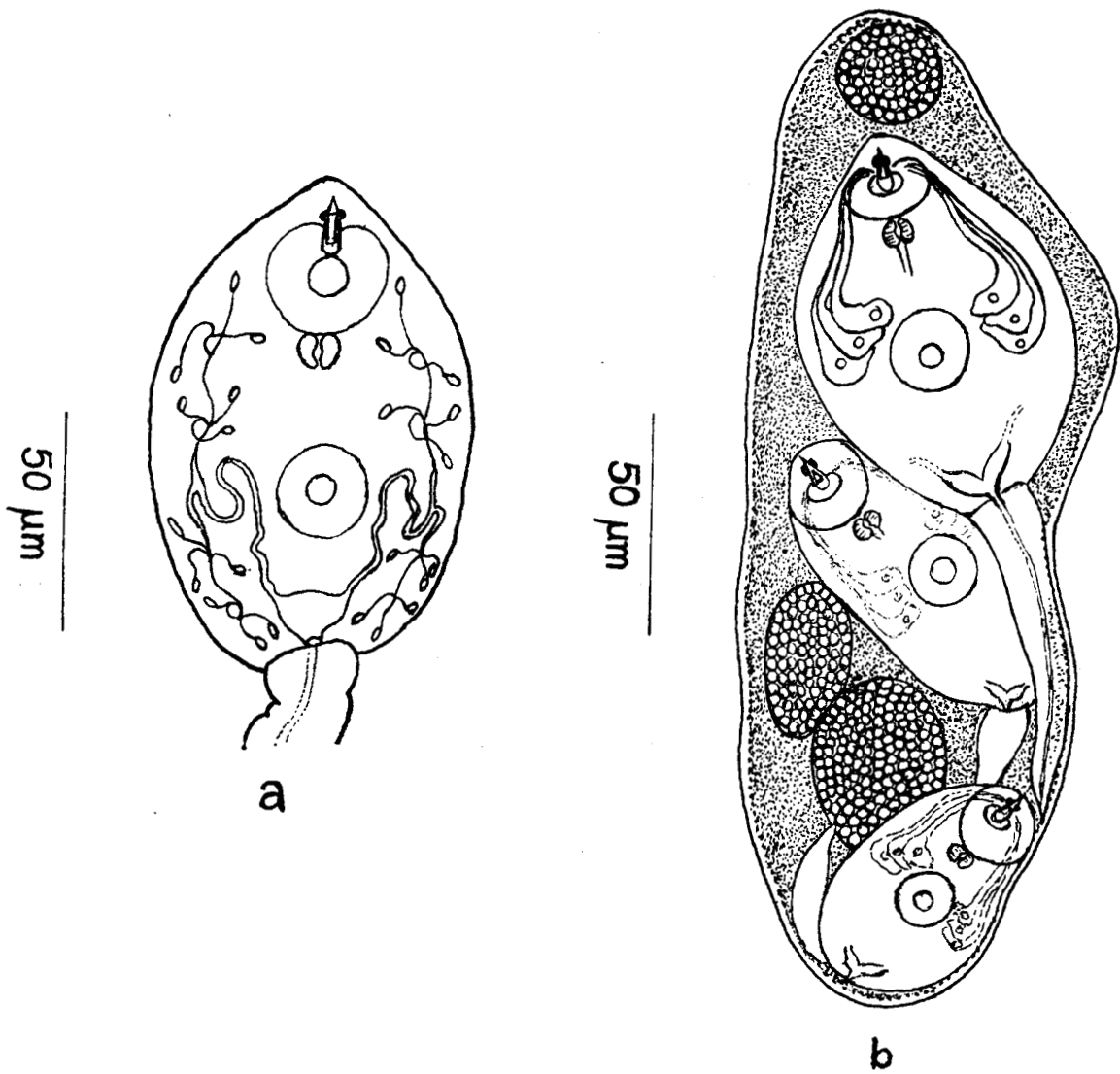


Fig. 28 : *Cercaria* sp. XI Malabar n. sp.
a. Excretory system; b. Sporocyst

Snail host	:	<i>Thiara tuberculata</i> (Mueller)
Site of infection	:	Hepatopancreas
Locality	:	Chaliyar in Malappuram district (Kerala)
Period of collection	:	November - December 2000
Prevalence	:	Five of 1161 (0.43%) snails examined were infected.

Remarks:

Among the non-virgulate xiphidiocercariae possessing 3 pairs of penetration glands, spinose body and aspinose tail, the present form comes close to *Cercariae indicae* XVIII Sewell, 1922, *C. indicae* XIX Sewell, 1922, *Cercaria leyteensis* no. 15 Ito, 1977, *C. melanopsis* I Ismail and Abdel-Hafez, 1983, *C. premvatai* Pandey and Singh, 1986, *C. baughi* Pandey and Singh, 1986, *C. srivastavi* Pandey and Singh, 1986 and *C. melanopsis* X Ismail and Blair, 1987.

However, it differs from the above cercariae in many respects. The present form could be distinguished from the *C. indicae* XVIII, *C. premvatai*, *C. baughi* and *C. srivastavi* by the structure of stylet and the absence of prepharynx. *C. indicae* XVIII disagrees further with the present form in the nature of penetration glands, and *C. premvatai*, *C. baughi* and *C. srivastavi* in having developed caeca also. The present cercaria deviates from *C. leyteensis* No. 15, *C. melanopsis* I and

C. melanopsis X in the structure and size of penetration glands, their contents and in the number of flame cells.

It is evident that the cercaria under report is different from the other related forms and therefore, it is reported here as a new species and named *Cercaria* sp. XI Malabar n.sp.

***Cercaria of Tremiorchis ranarum* Mehra and Negi, 1926**

(Fig. 29)

- Host : *Pila virens* (Lamarck, 1822)
- Locality : Kizhuparamba in Malappuram district.
- Period of collection : July 2001.
- Prevalence : Six out of 889 (0.67%) snails examined were infected.

Remarks:

The present xiphidiocercaria recovered from *Pila virens* has characters typical of cercaria of *Tremiorchis ranarum* as described by Rajendran and Janardanan (1993) and, therefore, identified and reported as cercaria of *T. ranarum*. Rajendran and Janardanan (1993) gave a detailed account of the life cycle of *T. ranarum* described by Mehra and Negi in 1926. They showed that the snail *Pila virens* acts as first intermediate host, tadpoles of *R. tigrina* and *R. cyanophlyctis*

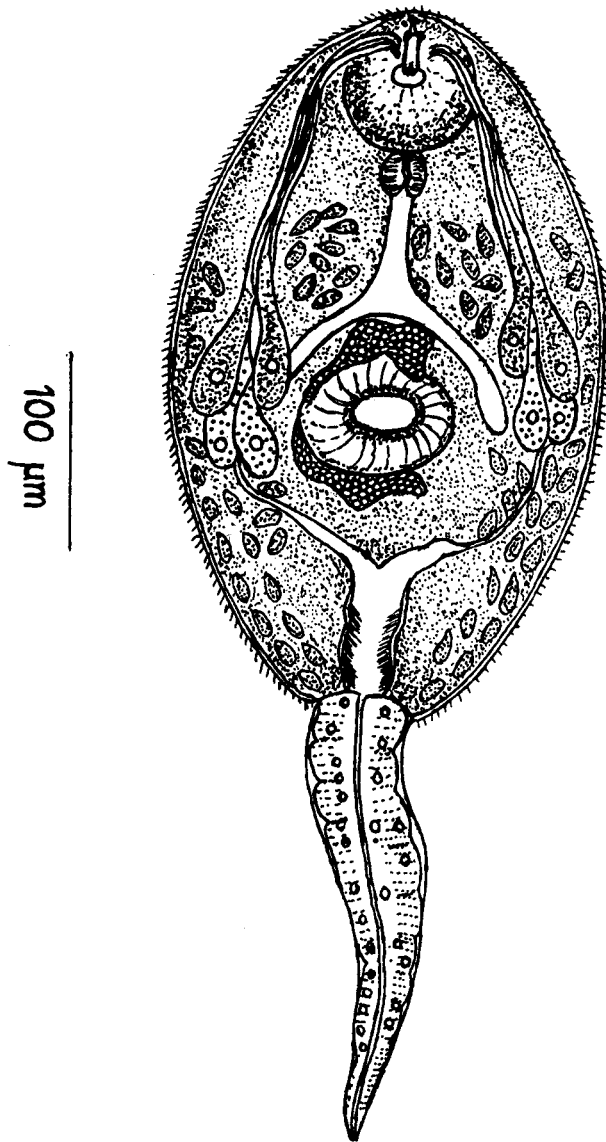


Fig. 29 : Cercaria of *Tremiorchis ranarum* Mehra and Negi, 1926

serve as second intermediate hosts and the frogs *R. tigrina* and *R. cyanophylctis* as the definitive hosts. This is the only available report on the life cycle of *T. ranarum*.

Cercaria of *Plagioporus panchax* n. sp.

- Host : *Paludomus transchauricus* (Gmelin)
- Locality : Kanhileri in Kannur district; Vattakundu in Wayanad district.
- Period of collection : September 2000 to November 2001.
- Prevalence : Fourteen of 1615 (0.86%) snails examined were infected.

Natural infections by the cercariae of *Plagioporus panchax* n. sp. were found in *P. transchauricus* collected from irrigation canals at Kanhileri in Kannur district and Vattakundu in Wayanad district. Metacercariae were recovered from the thoracic and abdominal muscles of stonefly naiads collected from the same localities. Adults were obtained experimentally in fishes, *Aplocheilus panchax*, *Rasbora daniconius* and *Danio aequipinnatus*, fed with 14-day-old metacercariae. Natural infections with adult flukes were found in the same fishes. The life cycle of *P. panchax* has been successfully established in the laboratory. An article on the

life cycle of *P. panchax* has been accepted for publication in *Rivista di Parassitologia* (3/2002, Vol. XIX). A copy of the paper is included under Appendix I.

Cercaria of *Centrocestus formosanus* (Nishigori, 1924)

Price, 1932

(Fig. 30)

- Host : *Thiara tuberculata* (Mueller)
- Locality : Padanakkad in Kasaragod district.
- Period of collection : October - November 2000.
- Prevalence : Four of 1161 (0.34%) snails examined were infected.

Natural infections by the metacercariae were found on gill filaments of *Danio aequipinnatus*, *Esomus danricus* and *Mystus malabaricus* collected from Padanakkad in Kasaragod district during November – December 2000.

Remarks:

The present cercaria recovered from *Thiara tuberculata* is identical with cercaria of *C. formosanus* described by Dhanumkumari *et al.* (1993b) from the same host species in Visakhapatnam. This cercaria was previously described by Sewell (1922) as *Cercariae indicae* III from *T. tuberculata* from Bombay and

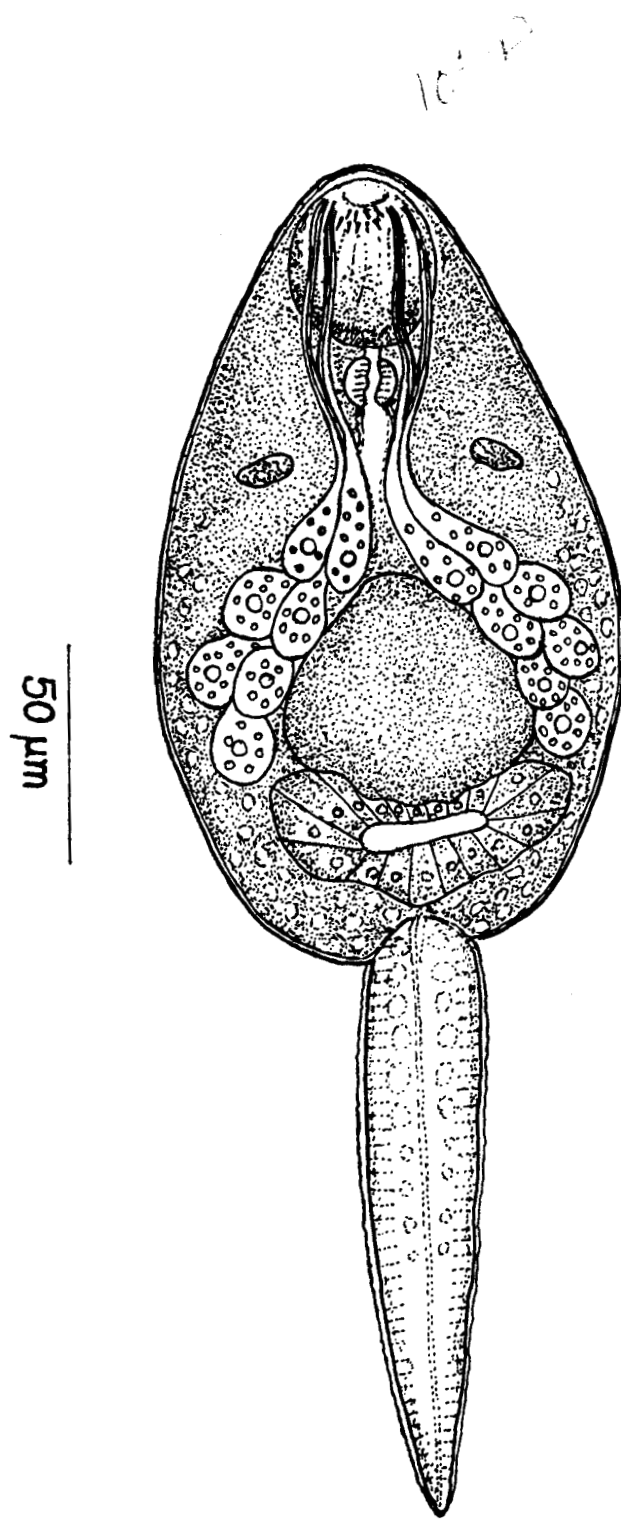


Fig. 30 : Cercaria of *Centrocestus formosanus* (Nishigori, 1924) Price, 1932

Calcutta and Ganapati and Rao (1983) described its behaviour. Chen (1942) and Martin (1958) reported *Melania tuberculatus chinensis* and *Stenomelania necombis* as first intermediate hosts for the fluke in China and Hawaii.

The life cycle of *C. formosanus* was first traced by Nishigori (1924) who showed that in Formosa the snail *Semisulcospira libertina* acts as the first intermediate host, 13 species of freshwater fish as second intermediate hosts and *Nyctcorax nyctcorax* as the definitive host. Subsequently, the life cycle of this parasite was worked out by Chen (1942, 1948) and Zhang *et al.* (1985) from China, Martin (1958) from Hawaii and Dhanumkumari *et al.* (1993b) from India.

The metacercariae of *C. formosanus* have been previously reported from the gills of freshwater fishes in India, namely, *Cirrhina reba*, *Amblypharyngodon mola*, *Hebeo bata*, *Esomus danricus*, *Puntius chola*, *P. sophore*, *P. ticto*, *Nandus nandus*, *Osteobrama cotio*, *Xenentodon cancilla*, *Channa punctatus*, *Notopterus notopterus*, *Chela laubuca*, *Oxygaster phulo*, *O. bacaila*, *Mastacembeles puncalus*, *Aplocheilus panchax* and *A. melastigma* (Nath and Pande, 1970; Pande and Shukla, 1972; Nath, 1974; Premvati and Pande, 1974; Madhavi, 1980a).

Recovery of the present cercarial species from *T. tuberculata* at Padanakkad in Kasaragod district forms a new geographical record.

Cercaria of *Haplorchoides mehrai* Pande and Shukla, 1976

(Fig. 31)

- Host : *Thiara tuberculata* (Mueller) and
Thiara scabra (Mueller)
- Locality : Padanakkad and Kanhangad in Kasaragod district;
Kanhileri in Kannur district; Azhinjilam in Malappuram
district, Malampuzha in Palakkad district.
- Period of collection : August to October 2000; November 2001.
- Prevalence : Sixteen of 1161 (1.38%) *T. tuberculata* and six of 178
(3.3%) *T. scabra* examined were infected.

Infections by this metacercaria were found in the freshwater fishes, *Puntius filamentosus*, *P. chola* and *P. ticto punctatus* collected from the above localities. The cysts were recovered from fin rays, base of caudal fin and beneath the scales.

Remarks:

This cercaria agrees fully with the cercaria of *Haplorchoides mehrai* Pande and Shukla, 1976, reported from *Melanoides tuberculatus* by Shameem and Madhavi (1988). Pande and Shukla (1976) reported the adult of *H. mehrai* from the cat fish, *Mystus vittatus* from Uttar Pradesh, and its metacercariae in fingerlings of 15 species of fishes. Shameem and Madhavi (1988) first demonstrated the life

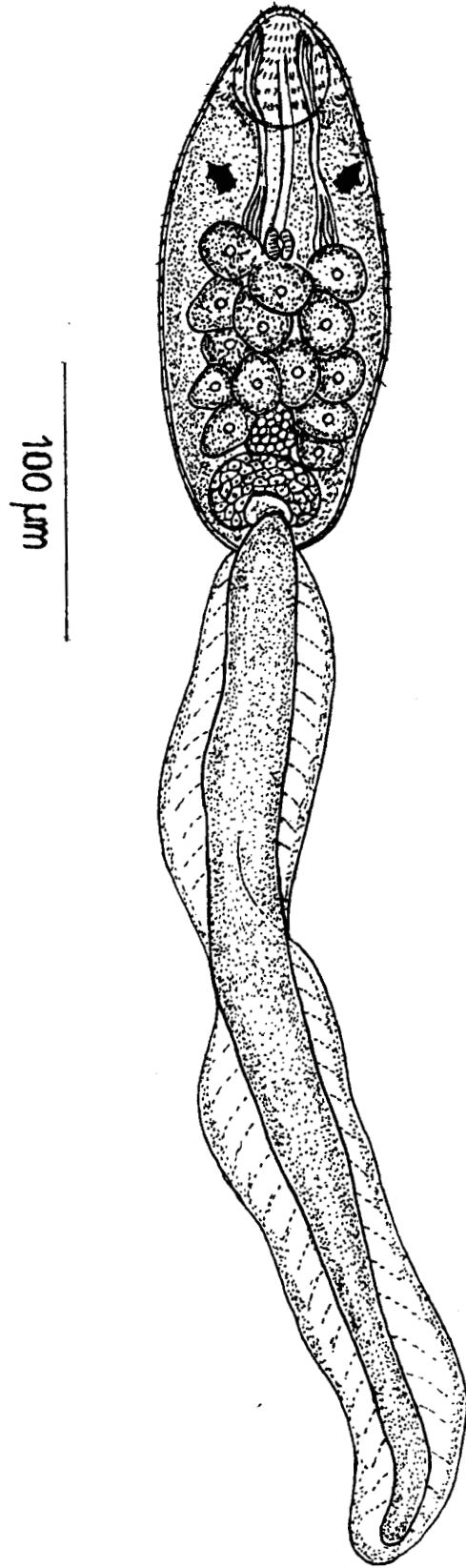


Fig. 31 : Cercaria of *Haplorchoides mehrai* Pande and Shukla, 1976

cycle of *H. mehrai* showing that the snail *Melanooides tuberculatus* acts as the first intermediate host, *Puntius sophore* as the second intermediate host and *Mystus gulio*, *M. vittatus*, *M. keleitus* and *Nangra robusta* as the definitive hosts in Chilka lake (Orissa). Recovery of the metacercaria from *P. filamentosus*, *P. chola* and *P. ticto punctatus* adds new host records for the metacercaria of *H. mehrai*. Occurrence of this cercaria in *T. tuberculata* and *T. scabra* of Kerala constitutes a new geographical record for this species.

Cercaria of *Acanthostomum burminis* (Bhalerao, 1926)

Bhalerao, 1936

(Fig. 32)

- Host : *Thiara tuberculata* (Mueller) and
Thiara scabra (Mueller).
- Locality : Neeleswaram and Padanakkad in Kasaragod district;
Kanhileri in Kannur district; Azhinjilam in Malappuram
district.
- Period of collection : February to April 2001; November 2001.
- Prevalence : Twenty-one of 1161 (1.81%) *T. tuberculata* and 2 of
178 (1.12%) *T. scabra* examined were infected.

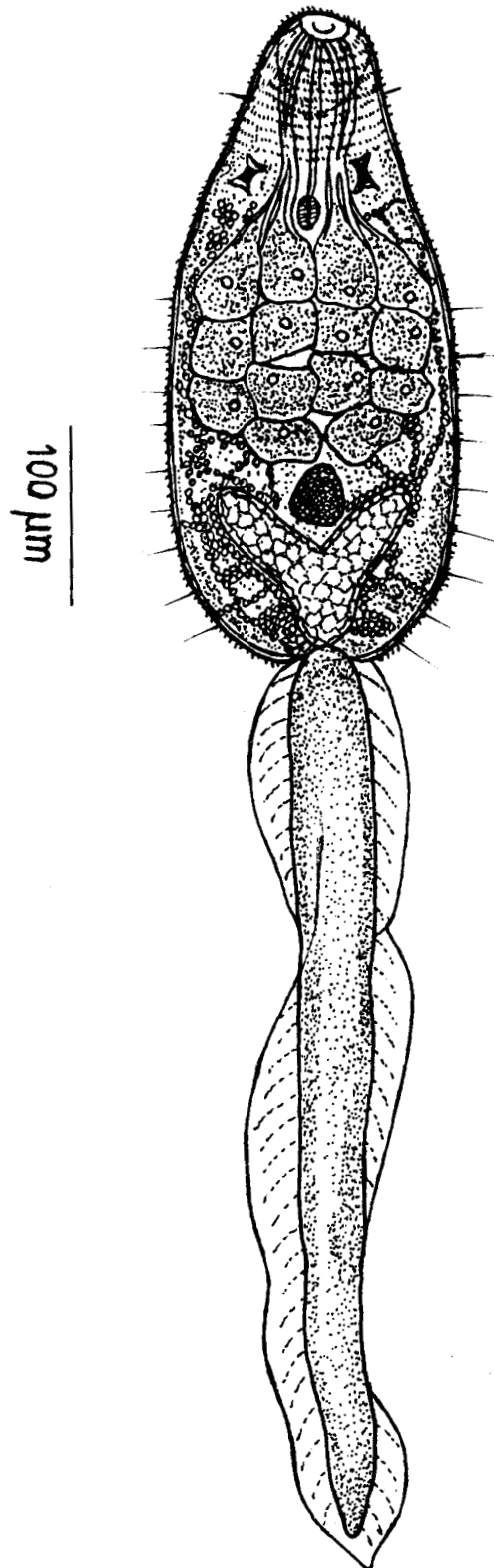


Fig. 32 : Cercaria of *Acanthostomum burminis* (Bhalerao, 1926)
Bhalerao, 1936

Rasbora daniconius and *Puntius parrah* collected from the above localities showed heavy infections with metacercariae of *Acanthostomum burminis*.

Remarks:

The cercaria can readily be identified as *Cercariae indicae* VIII Sewell, 1922 reported from *Melania tuberculata* of Calcutta, Bombay and Nilambur (in Kerala). Mohandas (1976) recorded the same cercaria from the same host in Palakkad, Ernakulam and Trivandrum districts of Kerala. Roopa and Janardanan (1998) elucidated the life cycle of *A. burminis* with *T. tuberculata* as the snail host, and recorded its metacercaria from fin rays of freshwater fishes like *Rasbora daniconius*, *Puntius parrah*, *P. melanampyx melanampyx*, *Mystus oculatus*, *M. malabaricus*, *Heteropneustus fossilis*, *Channa orientalis*, *Etroplus maculatus* and *Garra mullya*. The metacercariae developed into adults of *A. burminis* in the water snake *Xenochrophis piscator*.

A. burminis was originally described by Bhalerao (1926) from *Xenochrophis piscator* in Burma. The metacercariae of *A. burminis* have been previously reported from *Aplocheilichthys panchax*, *Mugil cephalus*, *Liza macrolepis* and *Valamugil cunnesius* in Andhra Pradesh (Madhavi, 1980a; Madhavi and Rekharani, 1985).

Recovery of this cercaria from *T. scabra* represents a new host record.

DISCUSSION

Vasandakumar M.V. “Studies on the larval trematodes (digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India” Thesis. Department of Zoology , University of Calicut, 2002

DISCUSSION

During the present investigation 10 species of freshwater snails representing 8 genera, collected from various localities of Malabar, were examined for cercarial infections. The collections were made during the period from February 1996 to December 2001. Of these, 6 snail species, *Pila virens* (Lamarck), *Thiara scabra* (Mueller), *T. tuberculata* (Mueller), *Paludomus transchauricus* (Gmelin), *Lymnaea luteola* Lamarck and *Indoplanorbis exustus* (Deshayes), were found to be infected by cercariae. The other 4 species, *Bellamya bengalensis* (Lamarck), *B. dissimilis* (Mueller), *Digoniostoma pulchella* (Benson) and *Gyraulus convexiusculus* (Hutton), were observed to be free from cercarial infection. In all, 22 species of cercariae, including 11 new species, were found in these snails. They belong to 8 categories, namely, furcocercous cercaria, amphistome cercaria, echinostome cercaria, macrocercous cercaria, gymnocephalous cercaria, xiphidiocercous cercaria, microcercous cercaria and pleurolophocercous cercaria. The new species of cercariae were recovered from 5 snail species. These include three furcocercous cercariae from *I. exustus*, one furcocercous cercaria from *L. luteola*, one furcocercous cercaria, one gymnocephalous cercaria and one xiphidiocercous cercaria from *T. tuberculata*, one echinostome cercaria from *P. virens*, and one macrocercous cercaria and two xiphidiocercous cercariae from *P. transchauricus*. Eleven known species of cercariae were obtained from 6 species of snails. The adults of these cercariae were known, and therefore, have been referred to by their

adult names. These are, cercariae of *Transversotrema patialensis* and *Centrocestus formosanus* from *T. tuberculata*, cercariae of *Diplostomum ketupanense* and *Cotylophoron indicum* from *I. exustus*, cercariae of *Fischoederius elongatus*, *Echinostoma revolutum* and *Petasiger variospinosus* from *L. luteola*, cercaria of *Tremiorchis ranarum* from *P. virens*, cercaria of *Plagioporus panchax* from *P. transchauricus*, and cercariae of *Haplorchoides mehrai* and *Acanthostomum burminis* from *T. scabra* and *T. tuberculata*. The species of cercariae recovered, their hosts and prevalence of infection are summarised in Table 21.

Life cycles of 2 species of trematodes have been elucidated during the present work using cercariae as the starting point. One is the life cycle of a new species of *Plagioporus*, *P. panchax*, an opecoelid trematode infecting the freshwater fishes, *Aplocheilus panchax*, *Rasbora daniconius* and *Danio aequipinnatus*. Natural infections with its cotylomicrocercous cercariae were found in *P. transchauricus*. These cercariae encysted in the musculature of stonefly naiads and became infective metacercariae in 14 days. An article on the life cycle of *P. panchax* has been accepted for publication in *Rivista di Parassitologia* (3/2002 Vol. XIX). A copy of the paper is included under Appendix I.

The other is the life cycle of *Petasiger variospinosus* (Odhner, 1910) Yamaguti, 1933, an echinostomatid fluke infecting the little cormorant, *Phalacrocorax niger*. Its 27-collar-spined cercariae emerged from *I. exustus* and encysted in the gill filaments of the freshwater fishes, *Esomus barbatus*,

Table 21. Summary of cercariae recovered, their hosts and prevalence of infection

Sl. No.	Cercaria	Host	No. of hosts		Prevalence (%)
			Infected	Examined	
1	2	3	4	5	6
1	<i>Cercaria</i> sp. I Malabar n.sp.	<i>Indoplanorbis exustus</i>	13	2545	0.51
2	<i>Cercaria</i> sp. II Malabar n.sp.	<i>I. exustus</i>	4	2545	0.16
3	<i>Cercaria</i> of <i>Transversotrema patialensis</i> (Soparkar, 1924) Crusz and Sathananthan, 1960	<i>Thiara tuberculata</i>	5	1161	0.43
4	<i>Cercaria</i> sp. III Malabar n.sp.	<i>Lymnaea luteola</i>	12	1943	0.62
5	<i>Cercaria</i> sp. IV Malabar n.sp.	<i>I. exustus</i>	9	2545	0.35
6	<i>Cercaria</i> of <i>Diplostomum ketupanense</i> Vidyarthi, 1937	<i>I. exustus</i>	11	2545	0.43
7	<i>Cercaria</i> sp. V Malabar n.sp.	<i>T. tuberculata</i>	3	1161	0.26
8	<i>Cercaria</i> of <i>Fischoederius elongatus</i> Rao and Ayyar, 1932	<i>L. luteola</i>	272	1943	13.9
9	<i>Cercaria</i> of <i>Cotylophoron indicum</i> Mukherjee, 1960	<i>I. exustus</i>	106	2545	4.17
10	<i>Cercaria</i> of <i>Echinostoma revolutum</i> (Frolich, 1802) Dietz, 1909	<i>L. luteola</i>	31	1943	1.6
11	<i>Cercaria</i> of <i>Petasiger variospinosus</i> (Odhner, 1910) Yamaguti, 1933	<i>I. exustus</i>	36	2545	1.41
12	<i>Cercaria</i> sp. VI Malabar n.sp.	<i>Pila virens</i>	9	889	1.01
13	<i>Cercaria</i> sp. VII Malabar n.sp.	<i>Paludomus transchauricus</i>	13	1615	0.8
14	<i>Cercaria</i> sp. VIII Malabar n.sp.	<i>T. tuberculata</i>	8	1161	0.68

1	2	3	4	5	6
15	<i>Cercaria</i> sp. IX Malabar n.sp.	<i>P. transchauricus</i>	6	1615	0.37
16	<i>Cercaria</i> sp. X Malabar n. sp.	<i>P. transchauricus</i>	8	1615	0.5
17	<i>Cercaria</i> sp. XI Malabar n.sp.	<i>T. tuberculata</i>	5	1161	0.43
18	<i>Cercaria</i> of <i>Tremiorchis ranarum</i> Mehra and Negi, 1926	<i>P. virens</i>	6	889	0.67
19	<i>Cercaria</i> of <i>Plagioporus panchax</i>	<i>P. transchauricus</i>	14	1615	0.87
20	<i>Cercaria</i> of <i>Centrocestus formosanus</i> (Nishigori, 1924) Price, 1932	<i>T. tuberculata</i>	4	1161	0.34
21	<i>Cercaria</i> of <i>Haplorchoides mehrai</i> Pande and Shukla, 1976	<i>T. scabra</i> <i>T. tuberculata</i>	6 16	178 1161	3.4 1.38
22	<i>Cercaria</i> of <i>Acanthostomum burminis</i> (Bhalerao, 1926) Bhalerao, 1936	<i>T. scabra</i> <i>T. tuberculata</i>	2 21	178 1161	1.12 1.81

Rasbora daniconius, *Puntius parrah*, *Mystus malabaricus* and *Aplocheilus panchax*. *P. niger* fed on fish containing infective metacercariae became infected. An article on the life cycle of this fluke has been accepted for publication in *Rivista di Parassitologia* (3/2002, Vol. XIX); a copy of which is included under Appendix II.

Observations were also made on further course of development of *Cercaria* sp. VI Malabar n. sp. infecting *P. virens*. This cercaria developed into metacercaria in the gills of the freshwater fishes, *Danio aequipinnatus*, *Labeo rohita*, *Aplocheilus lineatus* and *Macropodus cupanus*. The metacercaria developed 24 dorsally interrupted collar spines on a weakly developed collar. The metacercarial characters suggest of its relation to the genus *Microparyphium*. Attempts to develop the adult flukes in experimental hosts were unsuccessful. Natural definitive hosts are also not known.

Discussions on systematics of all the 22 cercarial species recovered during the present work have been made while describing them in the thesis. However, a general discussion on certain aspects of host-parasite relations of these cercariae is presented here.

Prevalence of infection

Of the 9347 snails examined only 620 were found infected by cercariae, indicating an overall prevalence of 6.6%. The prevalence of cercarial infection is

apparently not very high in the Malabar area. Comparable data on prevalence of infection were presented by several workers. Cort (1915) recorded that 8.5% of the molluscs of North America were infected. Dubois (1928) noticed that 28.8% of the snails in Switzerland were infected, whereas Kupriyanova-Shakhmatova (1957) reported 29.4% infection in the Volga region of the USSR and Zdarska (1963) noticed 23% in Czechoslovakia. In North Colorado the prevalence of infection was 35.5 (Acholonu, 1968), whereas in South Wales, it was 12 (Pike, 1968), in Philippines 4.78 (Ito *et al.*, 1977a), in the Kano area of Nigeria 16.6 (Ndifon and Umar-Yahaya, 1990) and in the Belo Horizonte region of Brazil it was 1.7 (Souza *et al.*, 1998).

Sewell (1922) while studying the cercarial fauna of India noted variability in the prevalence of infection from place to place. Mohandas (1974) reported that 10.23% of the snails in Kerala exhibited infection by 37 species of cercariae.

An interesting observation was that all the infected snail species were not equally preferred by the cercariae. *L. luteola* was the most preferred host with 16.2% of them infected by cercariae. Next to this was *I. exustus* with 7.03% infection, followed by *T. tuberculata* (5.3%), *T. scabra* (4.49%), *P. transcauricus* (2.53%) and *P. virens* (1.68%). The question as to why *L. luteola* is the most preferred host when compared to the other species is difficult to answer. It is an accepted fact that lymnaeid snails in general act as good intermediate hosts of digeneans. Probert (1966) and Mohandas (1974a) suggested that the family

Lymnaeidae may be phylogenetically very old and well adapted to withstand the effects of infection because of the long-term association between host and the parasite. The present results also support the view of these authors.

Another observation was that *T. tuberculata* is to be ranked first when the number of species of cercariae infecting each snail species in Malabar is taken into account. Of the 22 species of cercariae recorded, 7 were found in *T. tuberculata*, 6 in *I. exustus*, 4 in *P. transchauricus*, 3 in *L. luteola* and 2 in *P. virens*. Of these, the cercariae of *Haplorchoides mehrai* and *Acanthostomum burminis* recovered from *T. tuberculata* were also found in *T. scabra*. From an examination of the lists of cercariae known till date (Table 1 to 8) from the 8 genera of snails in India, it is apparent that the genus *Thiara* harboured the maximum number of cercarial species (72). Next to this was *Indoplanorbis* with 68 species, followed by *Lymnaea* (65 species), *Bellamya* (18 species), *Digoniostoma* (18 species), *Gyraulus* (11 species), *Pila* (5 species) and *Paludomus* (5 species). This data accumulated over the years is in general agreement with that collected during the present investigation. In either case, the genus *Thiara* shows the maximum number of species of cercariae. The reason why *T. tuberculata* is the most favoured host when the number of species of cercariae infecting it is considered is difficult to explain. The abundance of the snail cannot be offered as an explanation for this since other species of snails investigated are also more or less similar in abundance. Its wide-spread

distribution and ready accessibility to miracidial infection may be partly responsible for this situation.

The abundance of various groups of cercariae is presented in Table 22. Of the 22 species recovered, 7 belonged to furcocercous, 4 to xiphidiocercous, 3 each to echinostome and pleurolophocercous, 2 to amphistome and one each to gymnocephalous, macrocercous and microcercous cercariae. Although the furcocercous cercariae comprise 7 species, constituting about one third of the total cercariae recorded, they were found infecting only 57 snails as against 378 cases with 2 species of amphistome cercariae. The number of infected snails for four species of xiphidiocercous cercariae was 25, for 3 species of echinostome cercariae 76, for 3 species of pleurolophocercous cercariae 49, for one species of gymnocephalous cercaria 8, for one species of macrocercous cercaria 13 and for one species of microcercous cercaria 14. Wesenberg-Lund (1934) noted that furcocercous cercariae predominate in the lakes of Denmark. Probert (1966), Mohandas (1974a), Ndifon and Umar-Yahaya (1990), Choubisa (1991) and Toledo *et al.* (1998) also observed the predominance of furcocercous cercariae in the water bodies they studied. Amphistome cercariae proved to be numerically successful when compared to the other 7 groups of cercariae. The highest prevalence of amphistome infection is suggestive of the high incidence and intensity of *Fischoederius elongatus* and *Cotylophoron indicum* infections in ruminants of the region. This may also be the result of very efficient life cycles. It was also

Table 22. List of cercarial species recovered, their hosts and percentage of infection of the various groups in total snails infected

Sl. No.	Cercaria	Group	Snails						Percentage of groups in total snails infected
			<i>Pila virens</i>	<i>Thiara scabra</i>	<i>Thiara tuberculata</i>	<i>Paludomus transchauricus</i>	<i>Lymnaea luteola</i>	<i>Indoplanorbis exustus</i>	
1	2	3	4	5	6	7	8	9	10
1	<i>Cercaria</i> sp. I Malabar n. sp.							+	
2	<i>Cercaria</i> sp. II Malabar n. sp.							+	
3	<i>Cercaria</i> of <i>Transversotrema patialensis</i> (Soparkar, 1924) Crusz and Sathananthan, 1960				+				
4	<i>Cercaria</i> sp. III Malabar n. sp.	Furcocercous cercaria					+		9.19
5	<i>Cercaria</i> sp. IV Malabar n. sp.							+	
6	<i>Cercaria</i> of <i>Diplostomum ketupanense</i> Vidyarthi, 1937							+	
7	<i>Cercaria</i> sp. V Malabar n. sp.				+				
8	<i>Cercaria</i> of <i>Fischoederius elongatus</i> Rao and Ayyar, 1932						+		
9	<i>Cercaria</i> of <i>Cotylophoron indicum</i> Mukherjee, 1960	Amphistome cercaria						+	60.97

1	2	3	4	5	6	7	8	9	10
10	<i>Cercaria</i> of <i>Echinostoma revolutum</i> (Frolich, 1802) Dietz, 1909						+		
11	<i>Cercaria</i> of <i>Petasiger variospinosus</i> (Odhner, 1910) Yamaguti, 1933	Echinostome cercaria						+	12.26
12	<i>Cercaria</i> sp. VI Malabar n. sp.		+						
13	<i>Cercaria</i> sp. VII Malabar n. sp.	Macrocerous cercaria					+		2.1
14	<i>Cercaria</i> sp. VIII Malabar n. sp.	Gymnocephalous cercaria				+			1.29
15	<i>Cercaria</i> sp. IX Malabar n. sp.						+		
16	<i>Cercaria</i> sp. X Malabar n. sp.						+		
17	<i>Cercaria</i> sp. XI Malabar n. sp.	Xiphidiocercous cercaria				+			4.03
18	<i>Cercaria</i> of <i>Tremiorchis ranarum</i> Mehra and Negi, 1926		+						
19	<i>Cercaria</i> of <i>Plagioporus panchax</i>	Microcerous cercaria					+		2.26
20	<i>Cercaria</i> of <i>Centrocestus formosanus</i> (Nishigori, 1924) Price, 1932					+			
21	<i>Cercaria</i> of <i>Haplorchoides mehrai</i> Pande and Shukla, 1976	Pleurolophocercous cercaria			+	+			7.9
22	<i>Cercaria</i> of <i>Acanthostomum burminis</i> (Bhalerao, 1926) Bhalerao, 1936				+	+			

observed that the prevalence of infection varied from place to place and some water bodies were completely free from infection. As noted by Sewell (1922), we have only fragmentary information on the adult trematodes of this region or on their final hosts. Therefore, a satisfactory explanation cannot be offered regarding the variations in the prevalence of infection that are found to occur in different localities.

Host specificity

Strict host specificity was noted in 20 of 22 cercariae studied. *Cercaria* sp. I Malabar n. sp., *Cercaria* sp. II Malabar n. sp., *Cercaria* sp. IV Malabar n. sp., and cercariae of *Diplostomum ketupanense*, *Cotylophoron indicum* and of *Petasiger variospinosus* were found parasitizing only *I. exustus*; and cercaria of *Transversotrema patialensis*, *Cercaria* sp. V Malabar n. sp., *Cercaria* sp. VIII Malabar n. sp., *Cercaria* sp. XI Malabar n. sp. and cercaria of *Centrocestus formosanus* were found in *T. tuberculata* only. Similarly, *Cercaria* sp. III Malabar n. sp. and cercariae of *Fischoederius elongatus* and *Echinostoma revolutum* were found only in *L. luteola*; and *Cercaria* sp. VI Malabar n. sp. and cercaria of *Tremiorchis ranarum* in *Pila virens* only. *Cercaria* sp. VII Malabar n. sp., *Cercaria* sp. IX Malabar n. sp., *Cercaria* sp. X Malabar n. sp. and cercaria of *Plagioporus panchax* were found to infect only *P. transchauricus*. However, cercariae of 2 species, that of *Haplorchoides mehrai* and *Acanthostomum burminis*, were recovered from two hosts, *T. tuberculata* and *T. scabra*, showing that they

preferred more than one host. On these occasions the larvae were found in closely related snails of the same genus.

There are several examples in which one species of cercaria is found infecting either a single snail species or more than one. These types of specificity have been previously reported by several workers. Important among them are, Pike (1968), Nasir and Diaz (1973), Murty (1975b), Ito (1977a,b,c), Ito and Blas (1977, 1978), Mohandas (1977, 1981), Lengy and Gold (1978), Pandey and Agarwal (1978), Ramachandrula and Agarwal(1985), Choubisa and Sharma (1986), Choubisa (1991), Ramachandrula *et al.* (1992) and Ostrowski de Nunez *et al.* (1996).

During the course of the present investigation mixed infections involving 2 or more cercariae in a single snail host have not been observed. But examples of one host getting infected by more than one cercarial species are not rare. Such instances were reported by Lie *et al.* (1965), Probert (1966), Mukherjee (1966b), Pandey (1973a), Choubisa and Sharma (1986), Joudane *et al.* (1990), Ndifon and Umar-Yahaya (1990), Choubisa (1991), Ismail and Arif (1993), Makita *et al.* (1996), McCarthy (1999) and others. The factor(s) responsible for the different levels of host-specificity exhibited by cercariae are not understood. This aspect is open for future investigations.

SUMMARY

Vasandakumar M.V. “Studies on the larval trematodes (digenea) with emphasis on cercariae infecting freshwater snails in Kerala, India” Thesis. Department of Zoology , University of Calicut, 2002

S U M M A R Y

Ten species of freshwater snails belonging to 8 genera, collected from different water bodies in Kasaragod, Kannur, Wayanad, Kozhikode, Malappuram and Palakkad districts, during the period from February 1996 to December 2001, were investigated for their cercarial fauna. Of these, 6 species, *Pila virens*, *Thiara scabra*, *T. tuberculata*, *Paludomus transchauricus*, *Lymnaea luteola* and *Indoplanorbis exustus*, were found to be infected by larval digeneans. Altogether 22 species of cercariae, including 11 new species, have been recovered and studied. These belong to 8 categories: furcocercous cercariae (7 species), amphistome cercariae (2 species), echinostome cercariae (3 species), macrocercous cercaria (1 species), gymnocephalous cercaria (1 species), xiphidiocercous cercariae (4 species), microcercous cercaria (1 species) and pleurolophocercous cercariae (3 species). The eleven new species have been described in detail and designated as *Cercaria* sp. I to sp. XI Malabar series to indicate the region of collection. These include three furcocercous cercariae from *I. exustus*, one furcocercous cercaria from *L. luteola*, one furcocercous cercaria, one gymnocephalous cercaria and one xiphidiocercous cercaria from *T. tuberculata*, one echinostome cercaria from *P. virens*, and one macrocercous cercaria and two xiphidiocercous cercariae from *P. transchauricus*. Eleven known species of cercariae were obtained from 6 species of snails. The adults of these cercariae were known, and therefore, have been

referred to by their adult names. These are, cercariae of *Transversotrema patialensis* and *Centrocestus formosanus* from *T. tuberculata*, cercariae of *Diplostomum ketupanense* and *Cotylophoron indicum* from *I. exustus*, cercariae of *Fischoederius elongatus*, *Echinostoma revolutum* and *Petasiger variospinosus* from *L. luteola*, cercaria of *Tremiorchis ranarum* from *P. virens*, cercaria of *Plagioporus panchax* from *P. transchauricus*, and cercariae of *Haplorchoides mehrai* and *Acanthostomum burminis* from *T. scabra* and *T. tuberculata*. Intramolluscan stages of all these cercariae have been recovered and studied.

Life cycles of a new species of *Plagioporus*, *P. panchax*, and that of *Petasiger variospinosus* (Odhner, 1910) Yamaguti, 1933 have been successfully worked out during the course of present investigation. All the stages in the life cycles, from egg to egg-producing adults, have been established in the laboratory.

Observations were made on further course of development of a new echinostome cercaria, *Cercaria* sp. VI Malabar n. sp., infecting the apple snail, *Pila virens*. The cercariae exposed to the freshwater fishes, *Danio aequipinnatus*, *Labeo rohita*, *Aplocheilus lineatus* and *Macropodus cupanus*, developed into metacercariae. Experimental attempts to develop adult flukes were futile. The characters of cercaria and metacercaria indicate that in all probability these stages belong to the genus *Microparyphium*.

Data on prevalence of infection have been collected and presented. Host specificity and other aspects of host-parasite relations of cercariae infecting freshwater snails have been briefly discussed in the light of the available information.

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APPENDIX I

Rivista di Parassitologia
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Certificate

The document attests the fact that the following scientific articles:

- 1) The life cycle of *Plagioporus panchax* sp.nov. (Digenea: Opecoelidae) from freshwater fish in Kerala, India.
by **Vasandakumar M.V., Janardanan K.P.**
- 2) The life cycle of *Petasiger variospinosus* (Odhner, 1910) Yamaguti, 1933 (Digenea: Echinostomatidae)
by **Vasandakumar M.V., Janardanan K.P.**

have been accepted and their publication will be in *Rivista di Parassitologia* in issue 3/2002, Vol. XIX.



The Director
(Prof. Antonino Ioli)
A. Ioli

**THE LIFE CYCLE OF *PLAGIOPORUS PANCHAX* SP. NOV.
(DIGENEA: OPECOELIDAE) FROM FRESHWATER FISH IN
KERALA, INDIA**

M.V. VASANDAKUMAR K.P. JANARDANAN

The life cycle of *Plagioporus panchax* sp. nov., an opecoelid trematode of freshwater fishes, is elucidated and the life history stages are described. Natural infections with cotylomicrocercous cercariae were found in the freshwater snail, *Paludomus transchauricus* in Wayanad and Kannur districts of Kerala. Metacercariae were recovered from the musculature of stonefly naiads from the same localities. The course of development from cercaria to metacercaria was followed. Adults were obtained experimentally in fishes, *Aplocheilus panchax*, *Rasbora daniconius* and *Danio aequipinnatus*, fed with 14-day-old metacercariae. Natural infections with adult flukes were found in the same fishes.

INTRODUCTION

The Opecoelid genus *Plagioporus* Stafford, 1904 comprises a large number of species reported from freshwater and marine fishes from different geographical regions. There has been a number of erroneous interpretations made during the systematic history of the genus. Gibson and Bray (1982) revised the generic characters of *Plagioporus* and restricted it to freshwater forms only, and Shimazu (1990) and Gibson (1996) agreed with this reorganization. As far as is known, the

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genus at present has more than 27 species reported from freshwater fishes in different parts of the world. About 7 species have been reported under the genus *Plagioporus* by several workers in India. All these are from marine fishes, and therefore, should be considered as belonging to other genera.

So far life cycles of only 6 species of *Plagioporus* have been established. These are, *P. siliculus*, *P. angusticolle*, *P. sinitsini*, *P. shawi*, *P. hypentelii* and *P. skrajabini* (Sinitsin, 1931; Mathias, 1936, 1937; Dobrovolny, 1939; Schell, 1975; Hendrix, 1978; Chernogorenko *et al.*, 1978).

During an explorative study on the cercarial fauna of the freshwater snail, *Paludomus transchauricus*, collected from irrigation canals at Vattakundu in the Wayanad and Kanhilari in the Kannur districts of Kerala, we came across a cotylomicrocercous cercaria. Laboratory experiments with the cercaria revealed that it developed into a new species of *Plagioporus* in the freshwater fish *Aplocheilus panchax*, *Rasbora daniconius* and *Danio aequipinnatus* with stonefly naiads serving as second intermediate hosts. Natural infections with the adults were found in the intestines of the three fish species, and the metacercariae in the musculature of stonefly naiads collected from the same canals. The new species is named *P. panchax* and the present paper gives a full description of the adult and also its complete life cycle. This forms the first report of a *Plagioporus* sp. from freshwater fishes in India; it is also the first report of the life cycle of the genus from India.

MATERIALS AND METHODS

The snail *Paludomus transchauricus* were collected from irrigation canals of Vattakundu and Kanhilari from April 2000 to December 2001 and those infected with cotylomicrocercous cercariae were isolated. The cercariae were supravivally stained with neutral red and observed alive. Genital primordia were observed in lactoacetic carmine-stained cercariae. Measurements were made on heat-killed specimens. A few infected snails were later crushed and examined for intramolluscan stages of development.

To establish metacercarial infection experimentally, naiads of stoneflies obtained from an infection-free locality in Thusharagiri (Kozhikode District) were exposed for about 8 h to cercariae emerging from the infected snails. The stonefly naiads thus exposed were examined periodically to follow the course of development of the metacercariae. The metacercariae were recovered also from naturally infected hosts collected from the water bodies where infected snails were present. To obtain adult flukes experimentally, 14-day-old metacercarial cysts and those from natural infections of stonefly naiads were fed to clean fish, *A. panchax*, *R. daniconius* and *D. aequipinnatus* collected from snail-free man made ponds in the Calicut University Campus. The fed fishes were dissected at intervals, and the flukes at different stages of development were recovered. Flukes were recovered also from naturally infected fishes. Eggs liberated by mature flukes were collected

in filtered pond water and kept in cavity blocks at room temperature (26° – 28°C) to follow the development of the miracidium.

The metacercariae and adults were observed alive, using neutral red as vital stain. Those used for permanent preparations were fixed in AFA, and stained with Mayer's carmalum (Cantwell, 1981). The description of these stages are based on measurements of a minimum of 10 specimens. Measurements are given in micrometres; the range is followed by the mean value in paranthesis. Figures were made with the aid of a camera lucida and details added free hand from observations made on live specimens.

RESULTS

Laboratory and field studies demonstrated that the life cycle of *Plagioporus panchax* sp. nov. involves the snail *Paludomus transchauricus* as the first intermediate host, the stonefly naiads as the second intermediate host and the freshwater fish, *Aplocheilichthys panchax*, *Danio aequipinnatus* and *Rasbora daniconius* as the definitive hosts.

Egg (Fig. 1):

Eggs oval, yellow-brown, operculate, unembryonated, 46-77 (66) x 31-46 (41) in size.

Miracidium (Fig. 2):

Miracidia elongate oval or pyriform, 73-92 (84) x 16-28 (20) in size. Body covered with 4 tiers of ciliated epidermal plates in 6:7:4:2 pattern. Penetration glands 2, one on either side of the gut. Two flame cells present in midbody. Germinal mass, containing 6 to 10 nuclei, in posterior third of body.

Sporocyst (Fig. 3):

Sporocysts develop in the digestive gland of *P. transchauricus*. Sporocysts elongate, sausage-shaped, pale yellow, filled with 7 to 20 fully developed cercariae, and a few developing cercariae and germ balls; measured 893-1478 (1086) x 154-223 (168).

Cercaria (Figs. 4-5):

Natural infections by the cercariae were found in 14 of 696 (2.01%) *P. transchauricus*; infected snails were obtained only during September-November months. Cercariae emerged throughout night with peak emergence from 6 to 10 p.m. The cercariae performed leech-like movements typical of cotylomicrocercous cercaria. They attached to submerged objects by their adhesive tails and exhibited swaying movements with constant flexion and extension of body.

Description

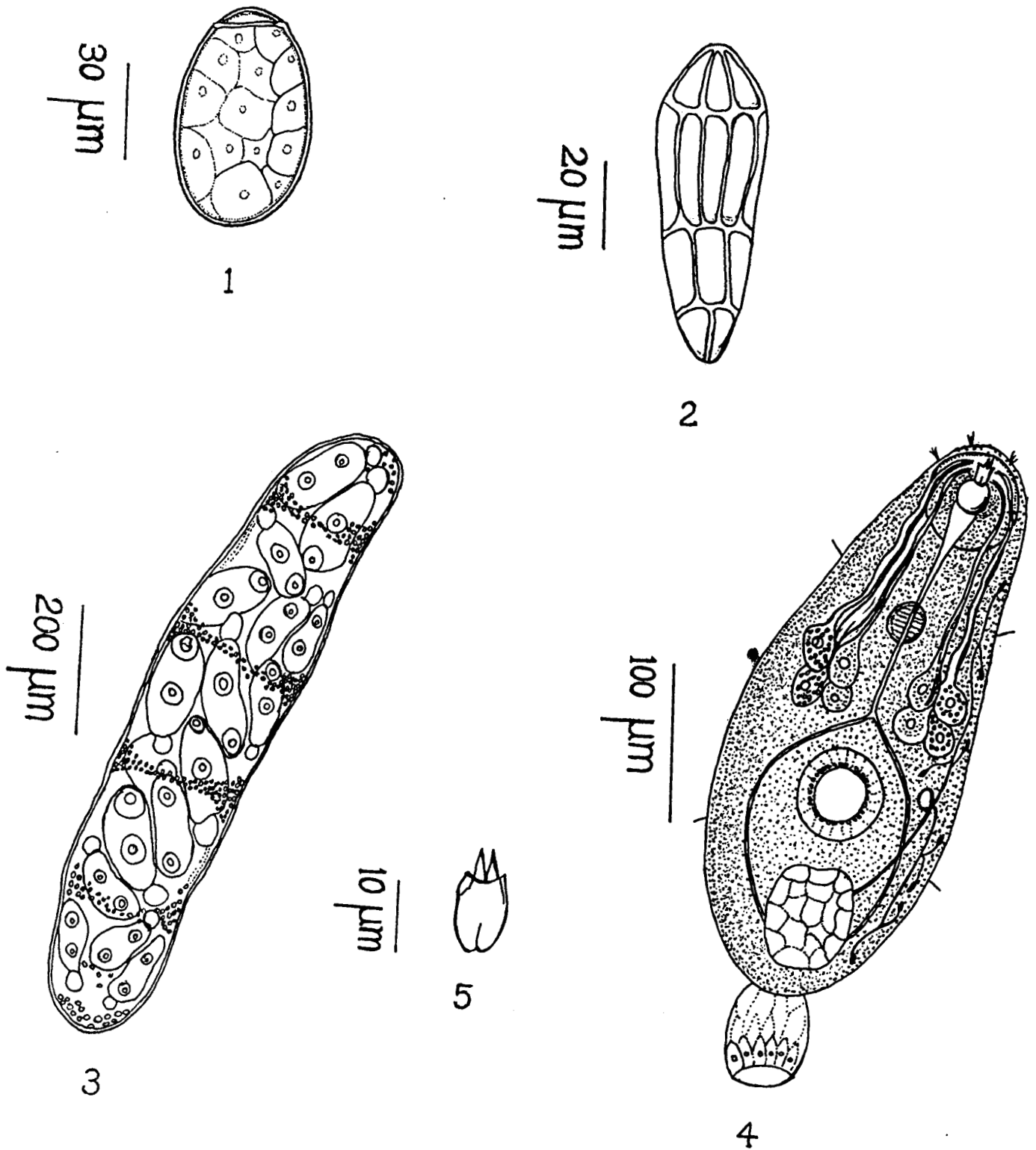
Cercaria cotylomicrocercous. Body elongate oval, aspinose, with 4 tuft of bristles at the oral sucker region and a few single ones scattered over the body;

measured 273-413 (331) x 89-160 (106). Tail cup-shaped, with glandular cells; 39-63 (49) x 33-53 (42) in size. Oral sucker roughly oval, with a few concentric circles of papillae in the inner margin; 46-56 (51) x 42-56 (47) in size. Stylet prominent, bifurcated in dorsoventral plane, directed dorsally; 13-17 (16) x 6-7 (6.4) in size. Acetabulum post-equatorial, protrusible, with concentric circles of papillae in the inner margin; 39-56 (47) x 42-56 (48) in size. Distance between oral sucker and acetabulum 89-154 (122).

Mouth sub-terminal, leads into a funnel-shaped buccal cavity. Pre-pharynx narrow, 33-76 (52) long. Pharynx muscular, globular, 16-23 (19) in diameter. Oesophagus moderately long; caecal bifurcation preacetabular; caeca extend to near posterior end of body.

Penetration glands 4 pairs, pre-acetabular; inner two pairs with finely granular contents, outer pairs with coarse granules. Ducts of penetration glands open individually near stylet shoulders.

Genital primordia consist of 'C'-shaped mass of cells in close association with acetabulum. Excretory bladder thick-walled, saccular and cellular, 39-73 (56) x 36-56 (48); main collecting ducts ascend from lateral walls of bladder and bifurcate to form anterior and posterior collecting ducts; excretory pore terminal; flame cell formula $2[(2+2) + (2+2)] = 16$.



Figs. 1-5: *Plagioporus panchax* sp. nov.
 1. Egg; 2. Miracidium with epidermal plate; 3. Sporocyst;
 4. Cercaria; 5. Stylet

Metacercaria (Fig. 6):

Natural infections with metacercariae were found in the thoracic and abdominal muscles of stonefly naiads. Prevalence of infection was 8% (3 of 36 stonefly naiads examined). Experimental infections have also been successfully established in clean stonefly naiads.

Cercariae penetrated the tegument of naiads within 2 to 3 h and completed encystment in 6 to 8 hours. One-day-old metacercariae were structurally similar to the cercariae except for the absence of penetration glands. Metacercariae were slightly larger, 168-188 (175) x 118-165 (139) in size, and without stylets when examined on the 4th day post-infection. On the 14th day post-infection, the cysts measured 231-241 (234) x 214-218 (215) in size, with larvae remaining folded inside the cyst cavity. These were infective and resembled those recovered from natural infections.

Description

Cysts yellow-brown, with an outer thick, opaque and an inner hyaline cyst wall. Body of metacercariae elongate oval, aspinose, 350-521 (429) x 132-156 (148) in size. Oral sucker subterminal, 46-58 (49) in diameter. Pre-pharynx short; pharynx 24-35 (33) x 24-28 (26). Oesophagus short; caeca extend to near posterior end of body. Primordia of ovary, testes and cirrus sac became conspicuous.

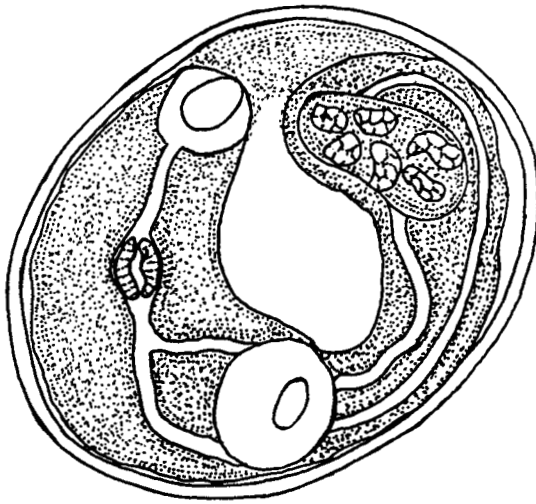
Adult (Fig. 7):

Natural infections with the adult flukes were observed in the intestine of 15 to 80 (19%) *A. pancrax*, 2 of 15 (13%) *R. daniconius* and 1 of 27 (13.7%) *D. aequipinnatus* collected from Vattakundu and Kanhilari in Wayanad and Kannur districts respectively. Experimental infections have also been successfully established in infection-free fish belonging to the 3 species.

Description

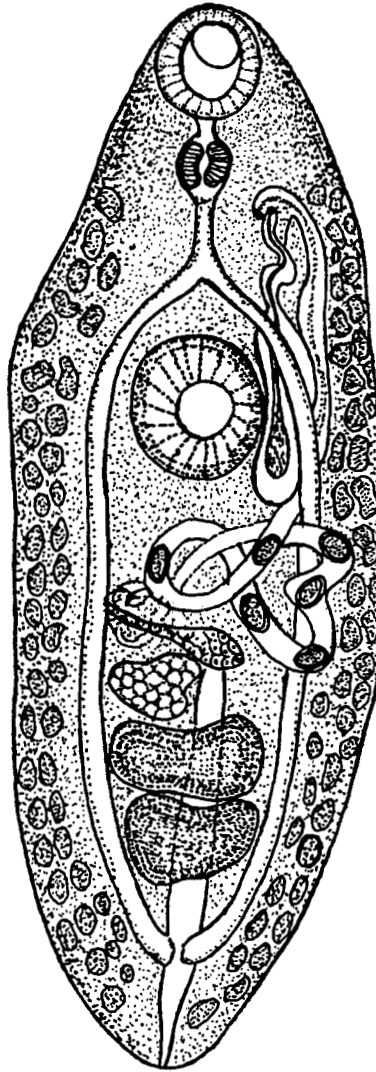
Body lanceolate, aspinose, measured 920-1448 (1054) x 296-524 (380). Oral sucker sub-terminal, 78-139 (103) x 78-139 (101) in size. Acetabulum larger than oral sucker in anterior part of middle third of body, 124-200 (155) x 117-200 (158) in size. Sucker width ratio 1:1.4 – 1.7 (1:1.5). Prepharynx short, 9-23 (17) long. Pharynx globular, 46-85 (65) x 38-85 (59). Oesophagus 62-146 (112) long; caecal bifurcation pre acetabular; caeca 577-1047 (751) long, narrow, terminating near posterior extremity and almost touching each other. Testes transversely oval, tandem, in posterior third of body; anterior testis 61-143 (101) x 77-262 (167), posterior testis 62-172 (120) x 140-246 (175). Cirrus sac measured 62-347 (214) x 31-85 (53), broad posteriorly, narrow anteriorly, and extending up to posterior margin of acetabulum; cirrus sac contains saccular seminal vesicle, pars prostatica and unarmed, eversible cirrus. Genital pore submedian, just anterior to caecal bifurcation. Ovary indistinctly trilobed, submedian, immediately anterior to testis;

60 μ m



6

300 μ m



7

Figs. 6-7 : *Plagioporus panchax* sp. nov.
6. Metacercaria; 7. Mature adult. 47

62-125 (92) x 53-203 (106) in size. Seminal receptacle and Laurer's canal present. Vitelline follicles numerous, extending laterally from posterior margin of pharynx to posterior end of body. Uterine coils between ovary and acetabulum, contain 10 to 19 eggs. Excretory bladder extended to the level of ovary; 124-322 (210) long.

- Holotype : Deposited in the Department of Zoology, University of Calicut, Kerala, India (No. Z/Par/Dig-2002-1)
- Type-host : *Aplocheilus panchax* (Hamilton) – Cyprinodontidae
- Additional hosts : *Rasbora daniconius* (Hamilton) – Cyprinidae
Danio aequipinnatus Mc Clelland – Cyprinidae
- Site of infection : Intestine
- Type locality : India, Kerala, Wayanad, Vattakundu
- Etymology : Named after the specific name of type-host, *A. panchax*.

DISCUSSION

The genus *Plagioporus* Stafford, 1904 belongs to the subfamily Plagioporinae Manter, 1947 of the family Opecoelidae Ozaki, 1925. Among the species of *Plagioporus* reported till date *P. shawi* (McIntosh, 1939) Margolis, 1972, *P. hypentelii* Hendrix, 1973, *P. sinitsini* Mueller, 1934 and *P. siliculus* Sinitsin, 1931 deserve comparison with the species under study. *P. shawi* resembles *P. panchax* sp. nov. in the extend of excretory bladder and cirrus sac, sucker ratios and

in having tandem testes and large post-testicular field, but differs in the distribution of vitellaria and nature of ovary. Further, cercaria of *P. shawi* is distinctly different in having simple, pointed stylet, 5 pairs of penetration glands, body surface without bristles and a different snail host. In the extend of vitellaria, position of genital pore and tandem distribution of testes, *P. hypentelii* and *P. sinitsini* come close to the present form. But in the extend of excretory bladder, cirrus sac and post-testicular field and sucker ratios they are different. Besides, the cercaria of the two species are also different in several respects. The present form is comparable to *P. siliculus* in the extend of excretory bladder and position of caecal ends. But, the latter has pre-acetabular cirrus sac, smooth ovary and its vitellaria extend only upto the level of caecal bifurcation. Cercarial characters of these two species are also different. Taking into consideration the differences of the present fluke with its closely related forms it would appear reasonable to treat the present fluke as a new species of the genus *Plagioporus*. We designate it as *P. panchax*, after the specific name of the type host, *A. panchax*.

Information on the life cycle of the genus *Plagioporus* is available for six species. Life cycle of *P. panchax* sp. nov. corresponds with that of the six species in that gastropod molluscs serve as first intermediate hosts, a wide range of freshwater arthropods are implicated as second intermediate hosts and adults occur in insectivorous fishes.

Cercaria of *P. panchax* sp. nov. recovered from *P. transchauricus* from Vattakundu in Wayanad and Kanhilari in Kannur, during the present investigation, differs from all known *Plagioporus* spp. cercariae reported from different parts of the world. But it agrees fully with *Cercariae indicae* XXXVIII Sewell, 1922 reported from the same species of snail collected from Arepetta Tea Estate in Wayanad in body organization, nature of stylet and in morphometry. But Sewell (1922) recorded only 3 pairs of penetration glands; in every possibility he might have overlooked a pair of penetration glands as it is very difficult to observe the fourth pair. Since the present cercaria and that described by Sewell are similar in organization and are infecting the same species of host in Wayanad, they are one and the same.

Natural infections with metacercariae of *P. panchax* sp. nov. were found only in stonefly naiads. Experimental infections also were established in stonefly naiads. It is an established fact that growth and differentiation of opecoelid metacercariae differ considerably from species to species. Cribb (1985) noted that in many species of the family Opecoelidae, the metacercariae continue to grow even after the infective stage is reached. This is, however, different in *P. panchax* sp. nov., where 14 days old infective metacercariae are of the same size as one month old larvae, and those obtained from natural infections of stonefly naiads did not attain a greater size.

Dobrovolny (1939) and Barger and Esch (2000) made some interesting observations on the life cycle of *P. sinitsini*. They reported that 3 different pathways exist in its life cycle pattern: a 3-host life cycle involving molluscan, arthropod and piscine hosts; a 2-host life cycle involving only molluscan and piscine hosts, in which the daughter sporocysts containing cercariae and metacercariae are voided from the alimentary canal of the snail *Elimia livescens* and the piscine hosts get infection by consuming the sporocysts; and a 1-host life cycle involving only the snail host. Here adults occur within daughter sporocysts voided with the faeces of the snail *E. symmetrica*, and these worms produced eggs containing active miracidia while still in the snail.

P. panchax sp. nov. has a 3-host life cycle. The cotylomicrocercous cercariae emerge from the snail, *P. transchauricus* and then encyst in the muscles of stonefly naiads. The metacercariae develop into adult flukes in the intestine of *A. panchax*, *R. daniconius* and *D. aequipinnatus*.

Although 14 *P. transchauricus* infected with cercariae of *P. panchax* were observed, metacercariae were not found developing inside the sporocysts and the sporocysts never came out with the faeces of infected snails. The phenomenon of progenesis was not observed in the metacercariae.

In the irrigation canals, where the life cycle of *P. panchax* occurs, the definitive and two intermediate hosts are quite abundant and, therefore, conditions

most congenial for successful completion of the life cycle occur within a closed ecological nich. The other 2 adaptive pathways found in the life cycle of *P. sinitsini* do not occur in this species as far as we could observe. The two truncated pathways have survival value and are adaptive, since these sustain the parasite's suprapopulation when the other host/hosts is/are not available in the habitat.

ACKNOWLEDGEMENT

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APPENDIX II

55

THE LIFE CYCLE OF *PETASIGER VARIOSPINOSUS*
(ODHNER, 1910) YAMAGUTI, 1933
(DIGENEA : ECHINOSTOMATIDAE)

M.V. VASANDAKUMAR K.P. JANARDANAN

The life cycle of *Petasiger variospinosus* (Odhner, 1910) Yamaguti, 1933 (Trematoda: Echinostomatidae) infecting the little cormorant, *Phalacrocorax niger* has been elucidated. Life cycle stages were successfully reared in the laboratory. Natural infections with 27-collar-spined echinostome cercariae were found in the planorbid snail, *Indoplanorbis exustus*. Metacercariae occurred in the gill filaments of 5 species of freshwater fishes. *P. niger* became infected when fed on fish containing infective metacercariae. The prepatent period is 21 days. Natural infections with adult flukes were found in the same birds collected from different localities in Malabar, Kerala.

INTRODUCTION

The genus *Petasiger* was established by Dietz, 1909 with *P. exaertus* from *Phalacrocorax carbo* as its type. Subsequently, 29 species have been added to the genus by several workers (Yamaguti, 1971; Ku *et al.*, 1977; Nassi, 1980; Kostadinova and Gibson, 1998). Of these, only 3 species have been reported from India: *P. nicolli* Pande, 1939; *P. yamaguti* Nigam, 1944 and *P. antigonus* Nigam,

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1944. But Mehra in 1980 synonymised *P. yamaguti* with *P. nicolli* making the number of valid species two. Life cycles of only 4 species have so far been elucidated. These are, *P. nitidus* by Beaver (1939), *P. australis* by Johnston and Angel (1941), *P. caribbensis* by Nassi (1980) and *P. grandivesicularis* by Kostadinova and Chipev (1992). The life cycle of an Indian species has not been established.

During an explorative study on the cercarial fauna of Malabar, we came across a 27-collar-spined echinostome cercaria in the planorbid snail, *Indoplanorbis exustus* collected from ponds and rivulets at Theerthankara in Kasargod, Palayad and Kanhilari in Kannur, Feroke and Ramanattukara in Kozhikode and Kizhuparamba and Iruvetty in Malappuram districts of Kerala. The cercariae encysted in the gill filaments of *Esomus barbatus*, *Rasbora daniconius*, *Puntius parrah*, *Mystus malabaricus* and *Aplocheilus lineatus*. The metacercariae fed to the little cormorant, *Phalacrocorax niger* developed into adults of the genus *Petasiger*. Among the 30 species of *Petasiger*, those with 27 collar spines include *P. exaeretus* Dietz, 1909, *P. variospinosus* (Odhner, 1910) Yamaguti, 1933, *P. nicolli* Pande, 1939 and *P. antigonus* Nigam, 1944. A comparative study revealed that the present fluke can readily be identified as *P. variospinosus* which was reported by Odhner (1910) as *Echinostomum variospinosum* from *P. auritus* in Egypt. Its complete life cycle has been worked out in the laboratory. Recovery of

this species from *P. niger* forms the first report of the species from India. It is also the first report of the life cycle of a *Petasiger* sp. in India.

MATERIALS AND METHODS

Indoplanorbis exustus were collected from different freshwater bodies in Malabar, Kerala during the course of 20 months, from May 2000 to December 2001. The snails were screened for cercarial infection and infected snails were isolated and kept in separate containers. The cercariae emerged from the infected snails were studied alive using vital stains. Collar spines of cercariae were studied following Lynch's (1933) silver impregnation technique. Study of excretory system was made by exposing cercariae to 0.01% urea solution. Genital primordia were observed in acetic orcein-stained cercariae. A few infected snails were later crushed and examined for intramolluscan stages of development. Measurements of cercariae were made on heat-killed specimens.

Infection-free *Esomus barbatus* and *Puntius parrah* were exposed to fresh cercariae emerging from snails to obtain metacercariae. Natural infections by metacercariae were found in fish collected from the waterbodies where infected snails were present. In order to obtain adults, the infected fishes were fed to *P. niger* free from parasites as determined by faecal investigations for the presence of eggs of *P. variospinosus*. The fed birds were dissected on 7, 10 and 21 days post-

infection to recover adults. Flukes were also recovered from naturally infected cormorants from Kannur and Malappuram districts.

The metacercariae and adults were studied alive, and those used for permanent preparations were fixed in AFA and stained with Mayer's carmalum, following the procedure outlined by Cantwell (1981). The eggs collected from faeces of infected birds were cleaned and kept in petridish containing well water for miracidial development. The emerged miracidia were studied alive. Lynch's (1933) technique was used to study the epidermal plates of miracidia.

Measurements are in micrometres (μm); the range is followed by mean values in parantheses. Illustrations were made with the aid of a camera lucida and details added free-hand from observations made on live specimens.

RESULTS

Egg and Miracidium (Figs. 1-2)

Eggs ovoid, yellow-brown, unembryonated, operculate, 93-156 (113) x 78-125 (103) in size. Eggs kept in well-water at room temperature (26-28°C) began to hatch in 9 days.

Miracidia small, elongate-oval, 96-113 (99) long and 40-50 (46) wide. Ciliated epidermal plates arranged in 4 rows, with 6, 6, 4 and 2 plates in each row from anterior to posterior end. Apical papilla present. Two lateral process at base of

each lateroanterior plates; primitive gut filled with refractile granules open at tip of apical papilla. A pair of pigmented eyespots posterior to gut. Two flame cells and a group of posteriorly placed germinal cells found in the body.

Redia (Figs. 3-4)

Mother rediae found in pericardial cavity of the snail host. Fully developed mother rediae motile, pale yellow, measured 610-820 (770) x 200-325 (270). Collar 210-360 (270) x 89-180 (156) in size. Mouth terminal; pharynx large, globular, muscular, 153-190 (170) in diameter. Gut saccular, extend upto the level of locomotor process.

Daughter rediae were found in the digestive gland. Fully developed rediae cylindrical, light orange, 1306-2063 (1850) x 385-690 (530) in size. Collar 200-320 (280) x 92-192 (176) in size; birth pore dorsal, posterior to pharynx. Mouth terminal; pharynx globular, muscular, 66-93 (79) in diameter. Gut extends up to three-fourth length of redia, filled with dark granules. Locomotor process well-developed, located near posterior end. Daughter rediae contained 2-10 cercariae at different stages of development and a few germ balls at psoterior end.

Cercaria (Fig. 5)

Natural infections with cercariae were found in 36 of 142 (25.35%) *Indoplanorbis exustus* collected from May 2000 to December 2001. Infected snails

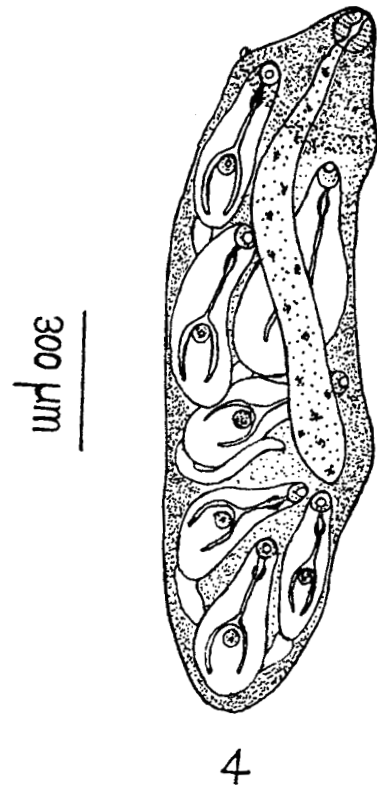
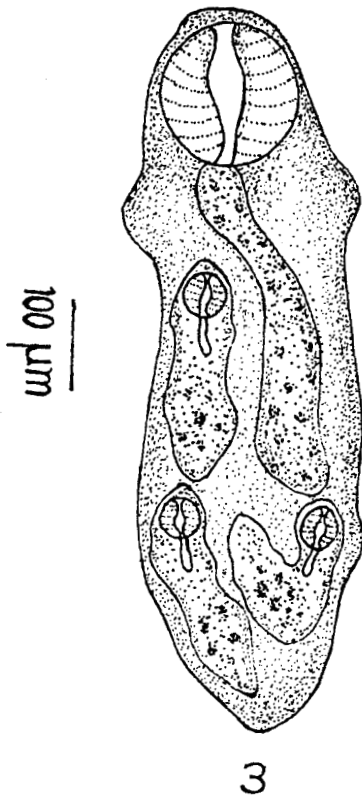
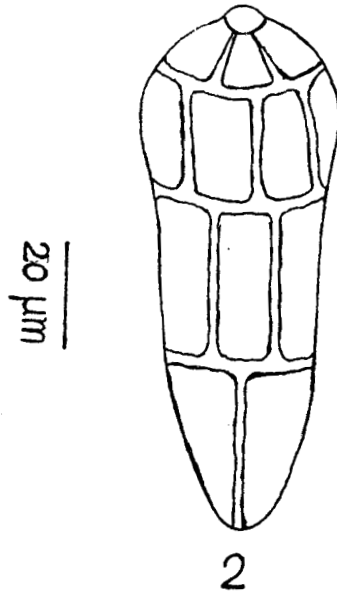
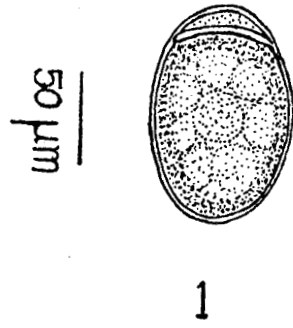
were present throughout the year, but infections were prevalent during October-February, when 39.5% of snails examined were positive for the cercaria.

Cercariae emerged from snails during day-time with peak emergence during morning hours. Cercariae were negatively phototactic, active swimmer and performed leech-like movements at the bottom of the container.

Description

Body elongate oval, aspinose, 315-460 (390) x 159-270 (198) in size, with 8-10 pairs of sensory hairs at anterior region. Collar distinct, 80-125 (110) wide, with 27 spines, 9-16 (13) in length. Arrangement of spines characteristic: 4 corner spines on each ventral lappet, 2 oral and 2 aboral; 5 laterals in single row on each side; 9 dorsals, 5 oral and 4 aboral. Oral sucker subterminal, circular, 40-60 (52) in diameter and opens into prepharyngeal sac with 2 large and several small granules. Acetabulum post-equatorial, protrusible, oval, 56-64 (59) x 63-92 (78) in size. Tail cylindrical, almost equal to body length, 254-450 (410) x 55-74 (67) in size; sensory hairs and finfolds absent.

Mouth subterminal. Prepharynx 19-35 (25) long; pharynx muscular, 27-39 (32) x 20-30 (26) in size. Oesophagus narrow, solid, 110-150 (132) long; consists of 8 cells with round nuclei and granular cytoplasm. Caecal bifurcation anterior to acetabulum. Caeca solid, 160-250 (210) long, extend to the level of excretory



Figs. 1-4 : *Petasiger variospinosus*
1. Egg; 2. Miracidium with epidermal plates; 3. Mother redia;
4. Daughter redia

bladder; each caecum consists of 12 cells with an additional cell at the point of bifurcation.

Penetration glands 6 pairs, along oesophagus with four pairs of duct openings on dorsal lip of oral sucker. Cystogenous glands numerous, filled with rod-shaped contents, distributed throughout body, but only a few in between oral sucker and pharynx. Genital primordia consist of two masses of cells, located anterior and posterior to acetabulum and connected by a chain of cells. Excretory system stenostomate; excretory bladder bipartite, the main collecting ducts distended between pharynx and acetabulum, each containing 20-30 refractile granules of almost uniform size. Caudal branch of excretory system opens at tip of tail. Flame cell formula $2[(2+2+2+2) + (1+1+2+2)] = 28$.

Metacercaria (Fig. 6)

Metacercariae were found attached to the base of gill filaments and gill arches of freshwater fishes, *Esomus barbatus* (5 of 16), *Rasbora daniconius* (2 of 8), *Puntius parrah* (4 of 20), *Mystus malabaricus* (3 of 8) and *Aplocheilus lineatus* (2 of 20). Experimental infections have also been successfully established in *Rasbora daniconius* and *Puntius parrah*. All the fishes exposed were infected.

Description

Cysts spherical to ovoid, 138-254 (195) x 145-240 (170) in size; cyst wall double-layered, with an outer, hyaline layer and inner, opaque layer. The larvae

remained folded inside the cyst cavity. Internal structures like collar spines, suckers, excretory concretions and caeca were visible through cyst wall.

Body elongate-oval, spinose, 214-360 (290) x 110-146 (125) in size. Collar distinct, 89-146 (108) wide, with 27 conspicuous spines. Collar spine arrangement as in cercaria. Oral sucker sub-terminal, 24-54 (37) x 33-46 (36) in size. Acetabulum post-equatorial, larger than oral sucker, measured 38-61 (49) x 34-60 (48). Prepharynx 15-30 (19) long; pharynx muscular, 29-49 (34) x 16-23 (20) in size. Oesophagus solid, long, measured 89-158 (121), consisting of 8 rectangular cells having distinct nuclei; bifurcates in front of acetabulum. Caeca extend to the posterior end, 153-286 (178) long. Genital primordia as in cercaria. Excretory bladder 'Y'-shaped, with 2 primary ducts filled with large excretory concretions extending up to pharynx.

Adult (Fig. 7)

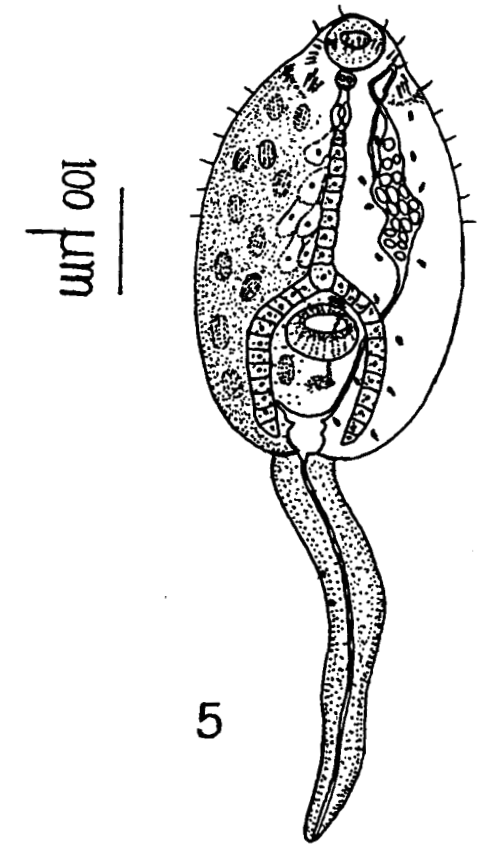
Natural infections with adult flukes were obtained from the duodenum and small intestine of the little cormorant, *Phalacrocorax niger* examined during the present study. All the birds collected from Kannur and Malappuram districts were infected. A single cormorant examined during December 2001 harboured as many as 32 flukes. Infections were successfully established in birds fed with infective metacercariae. It was found that metacercariae were infective when 18 days old and require 21 days to develop into mature adults in *P. niger*.

Description

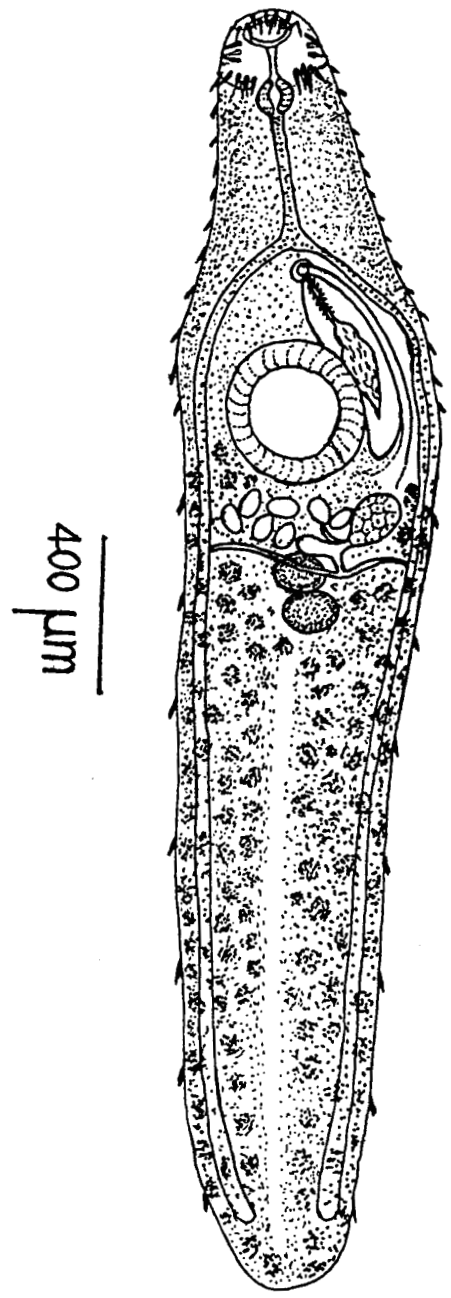
Body slender, elongate, measured 1560-3777 (2922) x 359-624 (508) in size. Body covered with scale-like spines up to about the middle of acetabulum, but sparsely distributed in post-acetabular region. Collar well-developed, 187-266 (239) x 280-359 (321) with 27 conspicuous spines, uninterrupted and alternating dorsally. Spine arrangement characteristic: 4 corner spines on each ventral lappet, 2 oral and 2 aboral, 86-110 (99) x 15.6-16.38 (15.69); 5 laterals on each side in single row, 62.4-93.6 (79.7) x 11.7; 9 dorsals, 5 oral and 4 aboral, 62.4-78 (75.4) x 11.7.

Oral sucker subterminal, 93-156 (113) x 78-124 (103) in size. Acetabulum larger than oral sucker, measured 343-468 (400) x 358-421 (383). Sucker width ratios 1:3.6. Pre-pharynx 40-79 (60) long. Pharynx ovoid, muscular, measured 93.6-109.2 (97) x 62.4-93.6 (80). Oesophagus long, slender, 312-624 (442). Caecal bifurcation just in front of acetabulum; caeca terminating near posterior extremity, 1950-2496 (2284) long.

Testes ovoid, smooth, post-acetabular, intercaecal, obliquely tandem, almost contiguous. Anterior testis 78-140 (105) x 93-110 (100), posterior testis 62-110 (83) x 93-125 (102). Cirrus sac large, elongate-oval, extends dorsally behind the posterior border of acetabulum; measured 287-490 (410) x 109-188 (143). Cirrus sac encloses single seminal vesicle, small pars prostatica, ductus ejaculatorius, and spined protrusible cirrus. Cirrus elongated, coiled, 234-250 (244) in length.



5



7



6

Figs. 5-7 : *Petasiger variospinosus*
5. Cercaria; 6. Metacercaria; 7. Adult

Ovary in midbody, spherical to ovoid, 98-156 (125) x 98-150 (110). Laurer's canal present. Uterus intercaecal, pre-testicular, containing 3-5 eggs. Seminal receptacle present. Vitelline follicles dark and granular, overlapping caeca in lateral fields, extending from posterior margin of acetabulum to posterior end of body, confluent in post-testicular region.

Discussion

The genus *Petasiger* Dietz, 1909 belongs to the subfamily Echinostomatinae (Looss, 1899) Faust, 1929. The adult flukes obtained naturally and experimentally from *Phalacrocorax niger* agree fully with the description of *Petasiger variospinosus* given by Odhner (1910) from *P. auritus* in Egypt and Bisseru (1957) from *Anhinga rufa levaillantii* in Zambia, and therefore, can readily be identified as *P. variospinosus*. This is the first report of *P. variospinosus* in India.

As far as is known, life cycle of an Indian species of *Petasiger* has not been worked out. The life cycle of *P. variospinosus* is established here for the first time. The present study demonstrated that cercariae of *P. variospinosus*, emerge from the planorbid snail, *Indoplanorbis exustus*, then encyst in the gill filaments of fish *Esomus barbatus*, *Rasbora daniconius*, *Puntius parrah*, *Mystus malabaricus* and *Aplocheilus lineatus* and become infective metacercariae in 18 days. *P. niger* serves as the definitive host where a 21 day prepatent period is spent by the fluke.

Four species of 27-collar-spined echinostome cercariae have so far been described. These are cercariae of *Paryphostomum radiatum* (Dujardin, 1845) Dietz, 1909 (as described by Johnston and Angel, 1942), *P. segregatum* Lie and Basch, 1967 (as described by Lie and Basch, 1967) and *Euparyphium melis* (Schrank, 1788) Beaver, 1941 (as described by Beaver, 1941) and *Cercaria unnaoensis* IV Pandey, Singh and Lal, 1982. The first three cercariae developed into species belonging to genera other than *Petasiger* and they differ from the present cercaria in several respects. *Cercaria unnaoensis* IV also differs from the present form in the number of corner spines, flame cells and penetration glands and in the absence of body setae. Therefore, the present cercaria is new, and is reported for the first time.

Metacercariae of *P. variospinosus* have been recovered from the gills of 5 fish species. Nath (1973) reported a similar metacercaria with 27 collar spines from *Channa punctatus* and *Rana cyanophlyctis* in Uttar Pradesh. A comparison of the present metacercaria with that described by Nath revealed that they are identical in most respects. Occurrence of the present metacercariae in *E. barbatus*, *R. daniconius*, *P. parrah*, *M. malabaricus* and *A. lineatus* in Malabar, Kerala constitutes new host and geographical record for this trematode.

In the tropical ponds and rivulets, where the life cycle of *P. variospinosus* is completed, the snail, fish and bird hosts occur in abundance throughout the year.

Here conditions most suitable for successful transmission and completion of the life cycle occur in a closed ecological nich. This explains the high prevalence of infections of the parasite found in the intermediate and definitive hosts. Experimental attempts to raise the adults in albino rats, one-day-old chicks and *Ardeola grayii* were futile. Adults develop in *P. niger* only as far as we could see. Thus *P. variospinosus* exhibits strict sepcificity at the level of definitive host.

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