A report on various digenetic metacercariae from the freshwater fishes of River Godavari, Rajahmundry

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Abstract Fishes serve as intermediate hosts to a number of avian digenetic trematodes. A survey on 10 different species of freshwater fishes belonging to 8 families from River Godavari from August 2005 to September 2007 revealed a total of 10 metacercariae of digenetic trematodes from five families i.e., Clinostomidae Lühe, 1901; Diplostomidae Poirier, 1886; Isoparorchidae (Travassos, 1922); Strigeidae Railliet, 1919 and Heterophyidae (Leiper, 1909) Odhner, 1914. The metacercarial fauna dominated the adult parasitic fauna of the freshwater fishes.

Keywords Metacercariae · Trematodes · Clinostomidae · Diplostomidae · Isoparorchidae · Strigeidae and heterophyidae

Introduction

Freshwater fishes offer a potential habitat to metacercariae of digenetic trematodes. Avian digenetic trematodes use fishes as secondary intermediate hosts to complete their life-cycles (Bullard and Overstreet 2008). These metacercariae inhabits every major organs of the host body like liver,

heart, gills, body cavity, etc. and cause severe pathologies to the body tissues (Rai and Pande 1969; Tort et al. 1987; Orecka-Grabda 1991; Watson et al. 1992; Laxma Reddy et al. 2006). Trematode metacercariae act as sentinel for changing limnological environment (Dzikowski et al. 2003) and are ecologically important. A pioneering amount of literature is available on the metacercariae of various digenetic trematodes from the fishes all over the world (Chandler 1951; Van der Kuyp 1953; Yamashita and Nishida 1955; Dollfus 1959; Hoffman 1960; Fischthal and Kuntz 1963; Ukoli 1966; Wootten 1973; Yamaguti 1975; Swennen et al. 1979; Font et al. 1984a; Britz et al. 1985; Kalantan et al. 1986, 1987; Malek and Mobedi 2001; Arafa et al. 2005 and Gustinelli et al. 2010). Also, considerable amount of work on digenetic metacercaria was contributed from India (Gupta 1961; Singh 1956; Khara 1958; Ganapati and Hanumantha Rao 1962; Rai and Pande 1965; Pandey 1966, 1971a, b, c, 1973; Thapar 1967; Chakrabarti 1970a, b, c, 1974; Chakrabarti and Baugh 1970; Agrawal 1975; Srivastava and Mukherjee 1976; Pandey and Agrawal 1978; Jain and Chandra 1979; Agrawal and Khan 1982; Gupta and Agarwal 1983a, b, 1984; Pandey and Tewari 1983; Tewari and Tyagi 1986; Sinha et al. 1988; Dhanum Kumari 1994; Jhansilakshmibai and Madhavi 1997 and Singh et al. 2003). In the present study, a total of ten species of metacercariae of five different families from 10 species of fishes of 8 families were described.

Materials and methods

A survey has been conducted to study the larval metazoan parasitic fauna from the fishes of River Godavari, Rajahmundry. 10 species of fishes belonging to 8 families were examined. A total of ten species of digenetic metacercariae

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belonging to 5 families were obtained (Table 1). Metacercarial stages of digenetic trematodes usually parasitize major organs of fish like oesophagus, gills, liver, heart and body cavity. Host body tissues were washed in physiological saline to remove excess mucus and were cut open with a longitudinal incision. Parasites were carefully alienated from the various body organs and collected in cavity blocks filled with saline solution. Larval trematodes were flattened between two slides or under slight pressure of slide and coverslip, post-fixed in AFA (Alcohol, Formalin and acetic acid in 85:10:5) and stained with alum carmine. Conventional techniques were employed for permanent whole mount preparations (Hiware et al. 2003; Madhavi et al. 2007). Figures were drawn with the aid of camera lucida and measurements are given in millimeters.

Results

A total of 10 species of metacercariae belonging to five families, Clinostomidae Lühe, 1901; Diplostomidae Poirier, 1886; Isoparorchidae (Travassos 1922); Strigeidae Railliet, 1919 and Heterophyidae (Leiper, 1909) Odhner, 1914 from ten species of fishes of Godavari River are described.

Metacercaria *Clinostomum dasi* Bhalerao, 1942 (Fig. 1-1)

Host. *Heteropneustes fossilis* Bloch

Habitat. Body muscles

No. of specimens. 3

Measurements Body $2.29\text{--}2.42 \times 0.65\text{--}0.71$; oral sucker $0.16\text{--}0.18 \times 0.20\text{--}0.23$; acetabulum $0.42\text{--}0.44 \times 0.38\text{--}0.39$; anterior testis $0.12\text{--}0.14 \times 0.16\text{--}0.18$; posterior testis $0.17\text{--}0.19 \times 0.21\text{--}0.23$; cirrus sac 0.40×0.08 ; ovary $0.07\text{--}0.08 \times 0.07\text{--}0.08$.

Remarks Leidy (1856) erected the genus *Clinostomum* with *C. gracile* as its type-species from fishes and frogs. There were reports of the metacercarial stages of different species of *Clinostomum* from fishes by Tubangui (1931), Bhalerao (1942), Jaiswal (1957), Kaw (1950), Paperna (1964) and Ukoli (1966) from many parts of the world. The metacercaria *C. dasi* was first described by Bhalerao (1942). Later, Pandey (1966), Jain and Chandra (1979) and Nama (1980) reported these parasites from *Heteropneustes fossilis*. Bhalerao (1942) described posterior testis as trilobed and the ovary as irregularly lobed which is immediately in front of the anterior testis. But in the present specimen, it is noted that posterior testis is irregularly lobed and the ovary

Table 1 List of metacercariae collected from the different families of freshwater fishes

S. no.	Name of metacercariae	Name of host	No. of hosts examined	Site of collection	No. of metacercariae obtained
1.	Fam: Clinostomidae Luhe, 1901	<i>Heteropneustes fossilis</i> Bloch	85	Body muscles	3
	1. <i>Clinostomum dasi</i> Bhalerao, 1942				
	2. Metacercaria <i>Clinostomum mastacembeli</i> Jaiswal, 1959	<i>Macrognathus aculeatus</i> Bloch	561	Body cavities	816
		<i>Mastacembelus pancalus</i> Hamilton	206	Oesophageal cavities	360
	3. <i>Clinostomum gideoni</i> Bhalerao 1942	<i>Barbus</i> sp.	85	Gills	4
	4. <i>Euclinostomum heterostomum</i> (Rud 1809) Travassos, 1928	<i>Channa punctatus</i> Bloch	252	Coelomic cavity and Liver	7
2.	Fam: Diplostomidae Poirier, 1806	<i>Belone cancila</i> Hamilton	185	Liver	145
	5. <i>Neascus hepatica</i> Chakrabarti, 1971				
	6. <i>Neascus</i> Hughes 1927 (<i>Neascus</i> Type-I)	<i>Anabas oligolepis</i> Bleeker	102	Gills	2
3.	Isoparorchidae (Travassos 1922)	<i>Mystus seenghala</i> Bloch	93	Air bladder	38
	7. <i>Isoparorchis hypselobagri</i> Billet, 1898				
4.	Strigeidae Railliet, 1919	<i>Glossogobius giurus</i> Hamilton	99	Body cavity	3
	8. <i>Tetracotyle glossogobi</i> Chakrabarti, 1970				
	9. <i>Tetracotyle</i> sp-I	<i>Macrognathus aculeatus</i> Bloch	561	Body cavities	1105
		<i>Mastacembelus armatus</i> Lacepede	494		31333
5.	Heterophyidae (Leiper, 1909) Odhner, 1914	<i>Macrognathus aculeatus</i> Bloch	561	Gills	242
	10. <i>Ascocotyle nana</i> Looss 1899				

Fig. 1 1 Metacercaria
Clinostomum dasi Bhalerao
1942; 2 Metacercaria
Clinostomum mastacembeli
Jaiswal 1957; 3 Metacercaria
Clinostomum gideoni Bhalerao
1942

is intertesticular in position and in this respect the parasite agrees with the descriptions of Pandey (1966). In spite of this variation, the present parasites are considered as *Clinostomum dasi* Bhalerao 1942.

Metacercaria *Clinostomum mastacembeli* Jaiswal 1957
(Fig. 1-2)

Host. *Macrognathus aculeatus* Bloch and *Mastacembelus pancalus* Hamilton

Habitat. Body cavity and Oesophageal cavities

No. of specimens. 816 and 360 parasites from *M. aculeatus* and *M. pancalus*

Measurements Body $4.95\text{--}10.86 \times 1.26\text{--}2.60$; suckers ratio $0.57\text{--}1.18$; oral sucker $0.23\text{--}0.31 \times 0.66$; acetabulum $0.63\text{--}0.84 \times 0.55\text{--}0.84$; testes $0.28\text{--}0.90 \times 0.23\text{--}0.81$; ovary rudimentary.

Remarks Jaiswal (1957) reported *Clinostomum mastacembeli* from *Mastacembelus armatus* from India. In the present study, parasites were found encysted heavily around the oesophagus in *Mastacembelus pancalus* and in the body cavity in *Macrognathus aculeatus*. The present parasites resemble *C. mastacembeli* in all respects except for a few variations in size and are considered as *Clinostomum mastacembeli* Jaiswal 1957.

Metacercaria *Clinostomum gideoni* Bhalerao 1942
(Fig. 1-3)

Host. *Barbus* sp.

Habitat. Gills

No. of specimens. 4

Measurements Body $2.57\text{--}3.20 \times 0.85\text{--}0.87$; oral sucker $0.17\text{--}0.26 \times 0.23\text{--}0.26$; Pharynx $0.13\text{--}0.14 \times 0.13\text{--}0.18$; acetabulum $0.40\text{--}0.53 \times 0.33\text{--}0.50$; anterior testis $0.25\text{--}0.26 \times 0.16\text{--}0.28$; posterior testis $0.17\text{--}0.22 \times 0.18\text{--}0.33$; ovary $0.10\text{--}0.12 \times 0.07\text{--}0.08$ and uterine sac $0.40\text{--}0.45$.

Remarks *Clinostomum gideoni* was first proposed by Bhalerao (1942) from the gills of the *Barbus sephere*. The parasite is characterized in having a ventrally bent anterior end, genital pore at the level of middle of anterior testis and characteristically lobed testes. In the present study the parasites are collected from the gills of *Barbus* sp. which resemble *C. gideoni* in all respects. Hence they are considered as *Clinostomum gideoni* Bhalerao 1942.

Metacercaria *Euclinostomum heterostomum* (Rudolphi, 1809) Travassos 1928 (Fig. 2-4)

Host. *Channa punctatus* Bloch

Habitat. Coelomic cavity and liver

No. of specimens. 7

Measurements Body $3.45\text{--}8.41 \times 1.28\text{--}2.36$; oral sucker $0.14\text{--}0.34 \times 0.20\text{--}0.52$; pre-pharynx $0.105\text{--}0.110$; pharynx $0.137\text{--}0.17 \times 0.16\text{--}0.44$; acetabulum $0.60\text{--}1.16 \times 0.55\text{--}1.34$; anterior testis U-shaped or slightly W-shaped, right arm $0.21\text{--}0.65$, left arm $0.22\text{--}0.63 \times 0.31\text{--}0.78$; posterior testis Y-shaped, right arm $0.14\text{--}0.38 \times 0.06\text{--}0.22$, left arm $0.07\text{--}0.39 \times 0.10\text{--}0.22$, base $0.08\text{--}0.26 \times 0.13\text{--}0.25$ and uterine sac $0.68\text{--}1.84 \times 0.110\text{--}0.57$.

Remarks The genus *Euclinostomum* was erected by Travassos (1928) with *E. heterostomum* as its type species from *Ardea purpurea* (type host), but later Van der Kuyp (1953) synonymized it with the genus *Tumaclinostomum*. However, Fischthal and Kuntz (1963) disagreed with Van der Kuyp and synonymized it with *Euclinostomum*. Several species of metacercaria of *Euclinostomum* Travassos 1928 were reported from India which include that of *Euclinostomum indicum* Bhalerao 1942 from *C. punctatus*; *E. channi* Jaiswal 1957 from *C. marulius* and *E. heptacaecum* Jaiswal 1957 from *C. punctatus*. Ukoli (1966) synonymized *E. bhagavantami*, *E. channi*, *E. heptacaecum* and *E. indicum* with *E. heterostomum*. Srivastava (1950) traced a part of the life-cycle of *E. heterostomum* but Donges (1974) gave a detailed account of its life-cycle and

agreed with Ukoli (1966) with regard to synonymy of various species. Sinha et al. (1988) reported *E. heterostomum* from the liver, spleen and kidney of *C. punctata*. Also, there were reports of distribution of *E. heterostomum* metacercarial infections in freshwater fishes, *Oreochromis mossambicus* by Britz et al. (1985). In the present study, the parasites resembles metacercariae of *E. heterostomum* (Rud 1809) Travassos 1928 in all characters.

Metacercaria *Neascus hepatica* Chakrabarti, 1971
(Fig. 2-5)

Host. *Belone (Xenentodon) cancila* Hamilton

Habitat. Liver

No. of specimens. 145

Measurements Body $1.18\text{--}1.34 \times 0.34\text{--}0.39$; forebody measures $0.74\text{--}0.79 \times 0.37\text{--}0.47$; hind body $0.53\text{--}0.58 \times 0.29\text{--}0.45$; oral sucker $0.03 \times 0.03\text{--}0.04$; acetabulum $0.04\text{--}0.05$; pharynx $0.01\text{--}0.02 \times 0.01\text{--}0.02$; hold fast organ $0.11\text{--}0.13 \times 0.14\text{--}0.21$; hold fast gland $0.04\text{--}0.06 \times 0.12\text{--}0.17$; Hind body with 3 well-defined, densely stained masses, two large and one small. Two large masses $0.09\text{--}0.10 \times 0.05\text{--}0.09$; posterior mass $0.13\text{--}0.16 \times 0.15\text{--}0.19$; ovary $0.06\text{--}0.08 \times 0.03\text{--}0.04$.

Remarks The larval genus *Neascus* was first erected by Hughes (1927) which includes eleven Indian species, *N. vetastai* Kaw 1950, *N. elongatus* Singh, 1957, *N. chelai* Khara 1958, *N. hepatica* Chakrabarti, 1971, *N. channi* Pandey, 1971, *N. xenentodoni* Pandey, 1971, *N. gussevi* Chakrabarti 1974, *N. hoffmani* Pandey, 1976, *N. komiyai* Pandey, 1976, *N. indicus* Thapar 1967 and *N. cirrhinus* Thapar 1967. Pandey (1971a) provided a key for the separation of *Neascus* type of larva reported from India. Chakrabarti (1970b) reported *N. hepatica* from *Xenentodon cancila*. The present parasites are collected from the same host and resemble *N. hepatica* in all characters except for the shape and size of body and development of pharynx. These differences may be considered as developing stage variations. So they are described as *Neascus hepatica* Chakrabarti 1971.

Metacercaria *Neascus* Hughes 1927 (Neascus Type-I)
(Fig. 2-6)

Host. *Anabas oligolepis* Bleeker

Habitat. Gills

No. of specimens. 2

Measurements Body $0.73\text{--}0.76$; forebody $0.57\text{--}0.59 \times 0.32\text{--}0.35$; pharynx $0.03\text{--}0.04 \times 0.02\text{--}0.03$; acetabulum

Fig. 2 4 Metacercaria
Euclinostomum heterostomum
 (Rud 1809) Travassos 1928; 5
 Metacercaria *Neascus hepatica*
 Chakrabarti 1971; 6
 Metacercaria *Neascus* Hughes
 1927 (*Neascus* Type-I)

0.04 in diameter; hold fast organ $0.10\text{--}0.11 \times 0.11\text{--}0.13$; hold fast organ 0.02×0.10 . Hindbody $0.19\text{--}0.20 \times 0.12\text{--}0.13$.

Remarks The larval genus *Neascus* was first erected by Hughes (1927). Quite a persuasive amount of work on the metacercaria of *Neascus* has been later contributed by Kaw (1950), Singh (1956), Khera (1958), Ganapati and Hanumantha Rao (1962), Trivedi (1964), Thapar (1967), Rai and Pande (1969), Pandey (1973) and Chakrabarti (1970b, 1974), Agrawal (1975), Pandey and Agarwal (1978), Agrawal and Khan (1982) and Dhanum Kumari (1994). The present parasites are immature and reproductive organs are not fully developed thus, could not be to identify up to species level and are hence considered as *Neascus* type-I.

Metacercaria *Isoparorchis hypselobagri* Billet, 1898 (Fig. 3-7)

Host. *Mystus seenghala* Bloch
 Habitat. Air bladders
 No. of specimens. 38

Measurements Body $2.66\text{--}3.68 \times 0.99\text{--}1.24$. Oral sucker $0.23\text{--}0.50 \times 0.10\text{--}0.29$. Pharynx $0.04\text{--}0.09 \times 0.04\text{--}0.14$. Acetabulum $0.39\text{--}0.57 \times 0.38\text{--}0.57$.

Remarks Metacercaria of *Isoparorchis hypselobagri* is associated with “Ink spot disease” in fishes. There are reports of this parasite in its immature and adult form by Bhalerao (1936), Wu (1938), Chauhan (1955); Dollfuss

Fig. 3 7 Metacercaria
Isoparorchis hypselobagri
Billet, 1898; 8 Metacercaria
Tetracotyle glossogobii
Chakrabarti, 1970; 9
Metacercaria *Tetracotyle* sp-I;
10 Metacercaria *Ascocotyle*
nana Looss 1899

(1959), Yamashita and Nishida (1955); Gupta (1961) and Rai and Pande (1965) from India, Japan, Australia, Java and China from a number of freshwater fishes. Rai and Pande (1965) reported juveniles of the parasite from *Mystus seenghala* and *Mystus vittatus*. Present parasites were collected from *M. seenghala* and resemble metacercaria of *Isoparorchis* in all characters, hence are described as Metacercaria of *Isoparorchis hypselobagri* Billet, 1898.

Tetracotyle glossogobii Chakrabarti, 1970 (Fig. 3-8)

Host. *Glossogobius giurus* Hamilton

Habitat. Body cavity

No. of specimens. 3

Measurements Body $0.50\text{--}0.74 \times 0.24\text{--}0.41$. Oral sucker $0.04\text{--}0.07 \times 0.06$. Pharynx $0.03\text{--}0.05 \times 0.03\text{--}0.04$. Oesophagus $0.07\text{--}0.10$. Ventral sucker $0.07\text{--}0.09 \times 0.09\text{--}0.10$. Pseudo-suckers $0.17\text{--}0.18 \times 0.07\text{--}0.08$. Tribocytic organ $0.15\text{--}0.23 \times 0.19\text{--}0.32$. Future ovary $0.04\text{--}0.05 \times 0.08\text{--}0.10$ testes $0.03\text{--}0.04 \times 0.10\text{--}0.11$.

Remarks *Tetracotyle glossogobii* was described by Chakrabarti (1970a) from the freshwater fish, *Glossogobius giurus* from India. About 18 species have been included under the larval genus *Tetracotyle* from India which include that of *T. ranae* Kaw 1950; *T. indicus* Singh 1956; *Tetracotyle* sp Rai and Pande 1969; *T. xenentodoni* Chakrabarti 1970; *T. muscularis* Chakrabarti 1970; *T. glossogobii* Chakrabarti 1970; *T. szidati* Chakrabarti and Baugh

1970; *T. lucknowensis* Pandey 1971; *T. Lali* Pandey 1971; *T. tandoni* Pandey 1973; *T. singhi* Pandey 1973; *T. baughi* Pandey 1973; *T. lymnaei* Pandey and Agrawal 1978; *T. gyanpurensis* Agrawal and Singh 1980; *T. sanjivi* Pandey and Tewari 1983; *T. fotedari*, *T. simhai* Pandey and Tewari 1983; and *T. satendari* Tewari and Tyagi 1986. The present parasites resemble *Tetracotyle glossogobii* in its body shape, size of pseudosuckers, size of holdfast organ and the host. Hence, the present parasites are considered as *Tetracotyle glossogobii* Chakrabarti 1970.

Tetracotyle sp- I. (Fig. 3-9)

Host. *Macrognathus aculeatus* Bloch and *Mastacembelus armatus* Lacepede

Habitat. Body cavity

No. of specimens. 1105 from *M. aculeatus* and 31333 from *M. armatus*

Measurements Body 0.57–0.72 × 0.34–0.42. Forebody 0.43–0.55 × 0.34–0.42, hindbody 0.14–0.18 × 0.23–0.26, Oral sucker 0.04–0.06 × 0.05–0.06, Pharynx 0.02–0.04 × 0.02–0.03, Ventral sucker 0.07–0.10. Pseudo-suckers 0.10–0.14 × 0.05–0.10.

Remarks Hoffman (1960) described the salient features of the various larval groups *Tetracotyle* Fillipi, 1859, *Neascus* Hughes 1927, *Prehemistomulum* Ciurea, 1923 and *Diplostomulum* Brandes, 1892. Quite an astounding work has been contributed on the *Tetracotyle* larval genus from freshwater fishes of India which include that of Kaw (1950), Singh (1956), Rai and Pande (1969), Chakrabarti (1970a, b, c), Chakrabarti and Baugh (1970), Pandey (1971b, c, 1973), Pandey and Agrawal (1978), Agrawal and Singh (1980), Pandey and Tewari (1983) and Tewari and Tyagi (1986). The present parasites were collected from *M. aculeatus* and *M. armatus* and could not be identified up to the species level and are named as *Tetracotyle* sp-I.

Metacercaria *Ascocotyle nana* Looss 1899 (Fig. 3-10)

Host. *Macrognathus aculeatus* Bloch and *Mastacembelus armatus* Lacepede

Habitat. Gills

No. of specimens. 242 from *M. aculeatus*; 452 from *M. armatus*

Measurements Body 0.28–0.32 × 0.05–0.07; Pre oral lobe 0.05–0.08; oral sucker 0.04–0.06; longer spines 0.015–0.020; small spines 0.007–0.010; additional spines 0.025–0.030; acetabulum 0.06–0.065; testes 0.018–0.023 × 0.15–0.020; ovary 0.010–0.015.

Remarks The genus *Ascocotyle* was first erected by Looss (1899). These heterophyid trematodes were commonly

found parasitizing the piscivorous birds and mammals, the metacercariae of which are found in fishes. Ransom (1920), Chandler (1941), Ostrowski de Núñez (1974, 1993), Font et al. (1984a, b), Salgado-Maldonado and Aguirre-Macedo (1991) reported these metacercariae from fishes. Its life-cycle studies were carried out by Leigh (1974), Ostrowski de Núñez (1993) and Scholz et al. (1997). In the present study, many parasites were collected from the gills of freshwater eels, *Mastacembelus armatus* and *Macrognathus aculeatus*. Scholz et al. (1997) also provided a key to the identification of *Ascocotyle* metacercariae. Present parasites come closer to *Ascocotyle nana* in many characters and hence are considered as metacercaria of *Ascocotyle nana* Looss 1899.

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