A Faunistic Survey of Cercariae from Fresh Water Snails: *Melanopsis* spp. and their Role in Disease Transmission

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Abstract

Snail transmitted diseases are one of the major group of helminth parasitic diseases which have been established by trematode parasites. The larvae of trematodes (cercariae) use the snails as host. The purpose of the present study was to identify of cercariae released from *Melanopsis* spp. (*M. doriae*, *M. costata*, *M. praemorsa*, and *M. nodosa*) and evaluate their medical importance. Accordingly, 2,266 *Melanopsis* spp. (fresh water snails) were collected from various agriculture canals in the central area of Khuzestan Province in the south west of Iran. 72 (3.1%) infected *Melanopsis* spp. snails were isolated and the cercariae were obtained by emerging or crushing methods. Subsequently, measurement and drawing were made on cercariae specimens and recognized. In some cases experimental infections were established in the animals for further identification. A total of 4 cercarial families and 1 cercarial group were identified as follows: Heterophyidae: *Haplorchis pumilio*, *H. taithui*, *Stellantchasmus falcatus* and *Centrocestus formosanus*; Echinostomatidae: *Echinocotyla*; Cyathocotylidae, Philophthalmidae and Monostome group cercariae (probably Notocotylidae). These results have been recorded for the first time and these cercariae are of medical and veterinary importance.

Keywords: *Cercariae*, *Melanopsis*, Iran

Introduction

Human schistosomiasis, fascioliasis, paragonimiasis, and clonorchiasis are very important snail transmitted diseases (1). Snails are the first intermediate host of trematode parasites. The larvae of trematodes (cercariae) use the snails as host. The purpose of the present study was to identify of cercariae released from *Melanopsis* spp. (*M. doriae*, *M. costata*, *M. praemorsa*, and *M. nodosa*) and evaluate their medical importance. Accordingly, 2,266 *Melanopsis* spp. (fresh water snails) were collected from various agriculture canals in the central area of Khuzestan Province in the south west of Iran. 72 (3.1%) infected *Melanopsis* spp. snails were isolated and the cercariae were obtained by emerging or crushing methods. Subsequently, measurement and drawing were made on cercariae specimens and recognized. In some cases experimental infections were established in the animals for further identification. A total of 4 cercarial families and 1 cercarial group were identified as follows: Heterophyidae: *Haplorchis pumilio*, *H. taithui*, *Stellantchasmus falcatus* and *Centrocestus formosanus*; Echinostomatidae: *Echinocotyla*; Cyathocotylidae, Philophthalmidae and Monostome group cercariae (probably Notocotylidae). These results have been recorded for the first time and these cercariae are of medical and veterinary importance.
Materials and Methods

The rural area in the north of Ahwaz city including, Bamdezh and Mazreeh districts of Khuzestan Province in the south west of Iran were chosen for this study, because there are so many canals there.

**Collection of cercariae from the infected snails**  
Melanopsis spp. snails particularly Melanopsis doriae were collected from the agriculture canals in the mentioned districts during 2000-2002. Snail samples were collected by a 1.5 meter long wooden handle paddle with a net size of 30 x 40 cm and transferred to Ahwaz Health Research Center. In this respect, 2,266 Melanopsis were examined for cercariae. Cercariae of Melanopsis snails were obtained by emerging or crushing methods. In the emerging method snails put in the Petri dish containing dechlorinated tap water and were placed against artificial light for two h or over night in the room. In the crushing method, snails were broken with tweezers and the soft tissues were placed between 2 slides and squashed. Collected cercariae were observed carefully as alive and then fixed in 50% ethanol or hot 5% formaldehyde and cleared in lactophenol or stained with azocarmin. Drawings were made by aid of a leica microscope drawing attachment (Camera lucida). Identification of cercariae species were made by using the systematic keys based on recognizable morphological characteristics or morphometric measurements (10, 11).

**Collection of metacercaria and adult parasites in vivo and in vitro situations**  
In some cases for developing cercariae to metacercariae or metacercariae to adult form of parasite, the laboratory animals including fish or rat and chicken were used, respectively. In case of heterophyid parasites, metacercariae were isolated from subscales, fins or gills of fishes (Gambosia affinis) which were placed in an aquarium near the infected Melanopsis snail. Adult forms of heterophyid trematodes were also collected from rats and chicken after oral infection via water containing metacercariae. For this purpose, intestinal tracts of rats and chicken were removed, dissected, scrap and searched carefully for collecting of adult parasites. Cyatocotylid metacercariae were obtained by gold fish oral infection and Philophtalmid metacercariae were collected from the bottom of the Petri dish shortly after emerging of cercariae from snail.

Results

**Frequencies of collected cercariae from Melanopsis snail**  
From the total of 2,266 Melanopsis spp. (Fig. 1), 72 (3.1%) snails were infected with various larval trematodes (cercariae). Cercaria species were as follows: Heterophyid cercariae (43 cases): Haplorchis pumilio, H. tathi, Stellantchasmus falcatus and Centrocestus formosanus; Echinostomatid cercariae (2 cases): Echinochasmus milvi; Cyathocotylid cercariae (19 cases) and Monostome group cercariae (probably Notocotylid trematodes) (3 cases) (Fig. 2-5). Heterophyid metacercariae and heterophyid adult parasites were obtained after one month from the infected hosts and cyatocotylid metacercariae isolated after 20 d from the infected Gold fish. Philophtalmid metacercariae were collected after 1-2 h emerging of cercariae from Melanopsis snails under stereoscope. Heterophyid cercariae had highest frequency, 59%, and echinostomatid cercariae had lowest frequency, 2%, among collected cercariae.

**Morphological specificity of collected cercariae**  
Cercariae of heterophyid trematodes belong to the parapleurolophocercous cercaria and have well developed finfold tail, vestigial ventral sucker and eyespots. In echinostomatid cercariae, the ventral sucker is in the mid-ventral surface of the body, oral sucker surrounded by a spiny collar and the cystogenous glands in the body are numerous. Cyathocotylid cercariae belong to vivax cercariae with vestigial ventral
sucker and have two pairs of longitudinal collecting ducts connected to excretory vesicle, dorso-ventral finfold on furcated tail. Philophthalmid cercariae belong to megalurus cercariae with adhesive gland cells in the tip of tail. Monostome group cercariae are without a ventral sucker and have 2-3 eye spots (10, 11).

Fig. 1: A. Melanopsis doriae snail, B. Melanopsis costata snail, C. Melanopsis praemorsa, D. Melanopsis nodosa snail from Khuzestan Province

Fig. 2: A. Heterophyidae metacercaria from infected fish, B. Heterophyidae cercaria from Melanopsis snail
Discussion

Heterophyid cercariae infect the local fish in the canals such as *Liza abu, Cyprinus carpi* and produce metacercariae in fish in the region (12). Infected fish cause heterophyiasis in the birds (13). Heterophyid trematodes as causative agent of heterophyiasis have been reported in these areas (9). Echinostomatid cercariae infect fish or snails as secondary intermediate hosts and birds or mammals including man as definitive hosts. Cyathocotyliid cercariae could infect fish as secondary host and birds or mammals as definitive hosts. Philophthalmid cercariae encyst and develop to adult in chicken or human eye (14). However, there are no reports on echinostomiasis and philohtalmiasis in the people of these districts. From the total of the cercariae species which are presented in this paper, heterophyid, echinostomatid, and philophthalmid cercariae have been reported from *Melanoides tuberculata* snail from Khuzestan province (15).

Various ecological factors, including season, water temperature, pH, oxygen and so forth are very effective on emerging of cercariae from the snails and their release inside water resources, where they could be penetrated into the secondary hosts (fish) such as heterophyid and echinostomatid cercariae or into the definitive hosts in the case of philophthalmid cercariae. For these reasons, during the hot months of May to Septem-
ber, when the local people (especially children) swim in the canals or ponds, philophthalmus cercariae will be able to encyst within the eye and could be a health problem in these areas. In addition, due to the increasing of amount cercarial shedding from snails in this period, cercariae transmission to fish is likely increased and uncooked fish may be serving as a source of heterophyiasis in humans in the region.

In summary, the presence of various cercariae of Melanopsis snails in the region and their potential for establishment of zoonotic diseases such as heterophyiasis, echinostomiasis and philophthalmiasis in man and animals may be concerned by health system authorities for surveillance of these fresh water snails.

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