

Original Article

**Prevalence of Cercariae Infection in *Lymnaea auricularia* (Linnaeus, 1758)
in NorthWest of Iran**

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Abstract

The pond snail *Lymnaea auricularia* serves as an intermediate host for many digenian species. In West Azerbaijan province, northwestern Iran, the cercariae infection investigation was undertaken in *L. auricularia* from May to November 2010. Of 6759 collected Lymnaeid snails, 370(5.5%) *L. auricularia* snails were identified. Cercariae infection was found in a number of 276 (74.56 %) snails. The results showed that removed cercariae from *L. auricularia* belonged to Echinostomercariae (96.38 %) and Furcocercariae (3.62 %) which were found in two out of 28 sites during the course of study. In summer and fall, the highest prevalence of cercariae infection was recorded for both identified cercariae. It is concluded that *L. auricularia* could be an important intermediate host of large group digenian trematodes in the region, which is necessary to take consideration in the control program of trematode infection.

Key words: Cercariae, *Lymnaea auricularia*, West Azerbaijan , Iran

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Introduction

Some freshwater snails (in particular *Basommatophora*) are intermediate hosts of parasitic flukes.¹ The flukes of the order Digenea have a complicated life cycle.² Freshwater snails are considered as the first and often also as the second intermediate hosts of parasitic trematode larva (cercariae), thus directly influence the distribution of the larva.^{2,3} Snail-transmitted diseases are one of the major groups of parasitic diseases which are established by trematode parasites. Therefore, the examination of the snails makes it possible to gain information about the sources of infection of definitive hosts, and also is the keystone for the elucidation of the trematode fauna in regions of interest.^{4,5}

Lymnaeidae (Pond snails) are freshwater gastropods which prefer inhabiting stagnant, slow streaming waters with heavy vegetation.⁶ They participate in the life cycle of at least 71 trematode species belonging to 13 different families. They parasitized birds and mammals as definitive hosts.⁷ In previous researches, the infection of Lymnaeid snails by larva of *Fasciola spp.*⁸⁻¹², *Paramphistomum spp.*¹³, *Holostom spp.*¹⁴ *Trichobilharzia spp.*¹⁵ have been reported. In Europe, Erasmus (1972) reported that several species of the genus *Lymnaea spp.* were infected by 29 different cercariae species.¹⁶ Sometimes, the same Lymnaeid species is used by more than one digenean species simultaneously.¹⁷ Forty species of *Lymnaea* snails were described¹⁷. In Iran, seven species of Lymnaeid snails were reported which *Lymnaea auricularia* (Eared pond snail) is one of the important pond snails.¹⁸ This freshwater snail is a aquaus dextral snail with a conical shell¹ which reported from a different parts of Iran like northwestern Iran.

So far, a few studies have been carried out on the abundance and diversity of cercariae infection in Lymnaeid snails, in particular *L. auricularia*, in Iran. For

instance, cercariae infection was reported in *L. gedrosiana* in Khoozestan province,^{19,20} *L. truncatula*²¹ in Khoozestan and Kurdistan, *L. gedrosiana*²² and *L. palustris*.²³ In West Azarbaijan province, no study has been undertaken on the abundance of cercariae infection in *L. auricularia* as yet. Thus, the objective of this study was to elucidate the diversity and regional distribution larval digenean trematode infection in *L. auricularia* of different parts of the province.

Material and Methods

Study area. Lymnaeid snails from 28 freshwater habitats including mountainous and low land areas of north, centre and south parts of West Azerbaijan province, northwestern Iran, were examined for cercariae infection from May to November 2010. The province has an altitude of 1,332 m above the sea level (ASL). According to Iranian Meteorological Organization (IMO) average temperature ranges from 9.4°C to 11.6°C (summer: 22.57°C and winter: 13.73°C) with mean annual rainfall of about 300-800mm (summer: 15mm and winter: 115.3mm) and relative humidity of 30 - 80 % (summer: 66 % and winter: 77.7 %). In general, this province has a semi-humid and temperate climate. Geographic Organization of Armed Forces (GOAF) indicated that there are numerous water bodies and reservoirs which together with relatively appropriate environmental factors provide favorable conditions for aquatic creatures, particularly for freshwater snails.

Collection of Lymnaeid snails and digenean cercariae identification. Lymnaeid snails were collected from wetlands, ponds, river basins, canals, springs, swamps, pools, streams, ditches and riversides by the same collector in each sampling for 15 min²⁴. The snails were put into a plastic screw capped containers containing the water of snail

habitat. Collected snails were transferred alive to the Malacology Lab for snail's specie identification and cercariae infection rate. *L. auricularia* was identified using key identification by Mansourian (1992) and Pflieger (1999).^{1,18} The snails were kept alive in an aquarium with optimal temperature and O₂ pressure to investigate cercariae infection.

To remove cercariae, *L. auricularia* snails were examined for the presence of released cercariae by shedding method.²⁵ The snails were individually placed in glass Petri-dish containing 50 ml dechlorinated tap water. Each Petri-dish was lit for 4-6 h with a 100-W light bulb at a distance of 15 cm so as not to increase water temperature and induce the expulsion of cercariae. If no cercariae shedding observed, snails were crushed under a stereomicroscope by forceps in order to find immature cercariae, sporocyst and/or redia. Collected cercariae or other stages were fixed in absolute ethanol (Merck, Germany) and stained with Azocarmin-Lactophenol.²¹ Stained cercariae were identified using systematic key by Frandsen and Christensen (1984).²⁶

Results

Lymnaeid snails. A total of 6759 was collected during the course of study. Of all snails, 370 (5.5 %) *L. auricularia* snails were identified and examined for the presence of cercariae (Fig.2).

Digenean cercaria infection. Cercariae infection was recorded in 74.59 % of

identified *L. auricularia* snails (Table1). The cercariae belonged to groups of larva including Echinostomercariae (96.38 %) and Furcocercariae (3.62 %) (Table 1, Fig.3). The cercariae infections were only recorded in larger (14-18 mm) and mature *L. auricularia* (Fig. 2). Echinostomercariae was found during the period of study (summer and fall). While cercariae infection rate was for Furcocercariae only in summer (Table1).

Geographical distribution. Infected snails were found in Golestaneh (83.65 %) and Zarrinehrud (19.23 %) in both mountainus and plain areas (Table 1, Fig. 1).



Fig 1. Map of the collected *L. auricularia* with cercariae infection in West Azerbaijan province, Iran (Go: Golestaneh, Zr: Zarrinehrud).

Table1. Prevalence and geographical distribution of cercariae infection in *L. auricularia* (N = 370) during the course of study in West Azerbaijan province, Iran.

Identified larva	Place			Time Season	Prevalence (%)	N	n
	Gd	Mo	Pl				
Furcocercariae	Zarrinehrud	-	+	Summer	32.26	31	10
				Fall	0	21	0
Echinostomercariae	Golestaneh	+	+	Summer	30.67	75	23
				Fall	100	243	243
Total						370	276

Gd: geographical distribution, Mo: Mountainus, Pl: Plain, N: number of examined snail, n: number of infected snail.



Fig 2. Shell of identified snail, *L. auricularia* (Dorsal and ventral position).

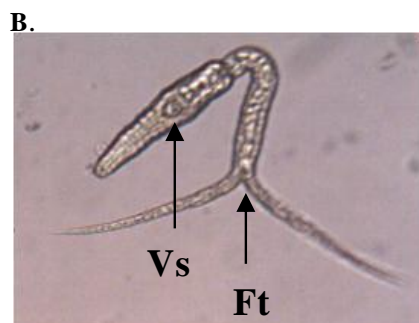
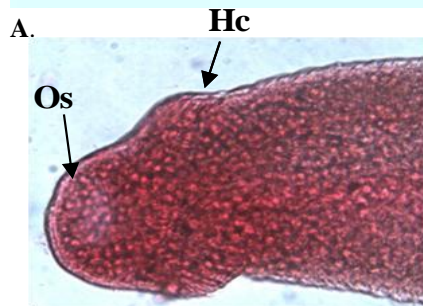
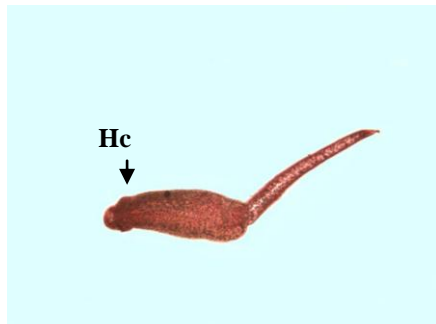


Fig 3. Echinostomecercariae (A, 100× and B, 400×) and Furcocercariae (C, 100×) removed from naturally infected *L. auricularia* (stained by Azocarmine-Lactophenol, Hc: Head collar, Os: Oral sucker, Ft: Forked tail, Vs: Ventral sucker).

Discussion

Intra-molluscan trematode parasitism is frequently associated with the alteration of a host's growth, fecundity or survival³¹. A considerable number of studies have attested the potential role of Lymnaeid snails in transmitting the infections by parasitic Trematodes worldwide.^{15, 27, 28, 29, 30,31}

In present study, *L. auricularia* snails were found in the province as previous investigation by Mansourian (1992).¹⁸ Cercariae infection in the snail was from two groups of fluke's cercariae (Furcocercariae and Echinostomecercariae). From different countries, infection of *L. auricularia* by cercariae such as Strigeidae, Schistosomatidae, Sanguinicilidae and Notocotylidae,¹⁵ *T. ocellata*³¹ and *T. franki*³² have been reported. In a recent investigation by Sharif *et al.* (2010) in Mazandaran province, 3.9 % of examined *L. gedrosiana* was infected by four cercariae groups.⁶ In Mazandaran province, cercariae infection by Echinostomecercariae was reported from *L. palustris* (1.22 %).³³ Nourpisheh (1998) noted that cercariae infection in *L. gedrosiana* of southwestern Iran was by four groups' fluke larva.²⁵ In Germany, *L. palustris* and *Radix (Lymnaea) auricularia* have been introduced as the intermediate host of Echinostomecercariae by Faltynkova and Haas (2006).¹⁵ In general, the presence of digenian larva stages of trematodes in *L. auricularia* indicates the important role of the snail in transmission of parasites at least to aquatic wild birds and probably to the domesticated birds in the studied area.

In this study, the cercariae infection rate was determined in summer and fall. The infection was found in wetlands where various types of wild aquatic-birds were living in. The reason may be due to favorable climatic conditions in the area make it attractive for the birds to migrate and settle during mid spring to late

summer. Thus, a cyclic host-parasite interaction could occur. Sharif *et al.* (2010) reported highly cercariae infection rate was between August-September.⁶ The Majority infection rate by schistosome larvae of birds was recorded between May-October.³⁶ According to Horak *et al.* (2002), the larvae released in spring constitute a source of infection for their definitive hosts returning from their wintering sites.³⁴ Therefore, the first cercariae developing after the spring infestations of snails could not appear before the presence of the birds. On the other hand, there is a possibility that snails infected by bird's larval trematodes can live throughout the winter by hibernation. This phenomenon was supported by the results of studies on the behavior of *L. stagnalis* in response to temperature.³⁵

In conclusion, this is the first step toward providing data on the distribution of parasitic trematodes infection of snails and their potential as intermediate host in the region. This study may provide a valuable basis for designing and launching an all-round control programmed in this area.

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References

1. Pflieger V. A field guide in colour to mollusks. Aventinum Nakladatelstvi, S.T.O., Polygrafia, Czech Republic, Prague, 1999; pp.: 28-29.
2. Esch GW, Curtis LA, Barger MA. A perspective on the ecology of trematode communities in snails. *Parasitol* 2001; 123: 57-75.
3. Farahnak A, Vafaie-Darian R, Mobedi I. A faunistic survey of cercariae from fresh water snails: *Melanopsis* spp. and their role in disease transmission. *Iranian J Publ Health* 2006; 35(4):70-74.
4. Bock D. Cercarien und Parthenitae (trematoda) aus Sußwasserschnecken des Naturreservates Obedska Bara bei Belgrad (Jugoslawien). *Zoologische Jahrbucher. Abteilung fur Systematik* 1982; 109:211-267. (in German)
5. Vojtek J. The importance of life-history studies for the systematics of the genus *Apatemon* (Trematoda, Strigeidae). In: *Parasitic worms and aquatic conditions* (Eds. R. Ergens and B. Rysavy). Proceedings of a symposium held in Prague, 1962; pp.:121-130.
6. Sharif M, Daryani A, Karimi SA. A faunistic survey of cercariae isolated from lymnaeid snails in central areas Mazandaran, Iran. *Pakistan J Biol Sci* 2010; 13(4):158-163.
7. Brown DS. Pulmonate molluscs as intermediate hosts for digenetic trematodes. In: Fretter, V., Peake, J. (Eds.), *Pulmonates. Systematics, Evolution and Ecology*, Vol. 2A. Academic Press, London, 1978; pp.:287-333.
8. Bargues MD, Artigas P, Meraysierra RL, et al. Characterization of *Lymnaea cubensis*, *L. viatrix* and *L. neotropican* sp. the main vectors of *Fasciola hepatica* in Latin America, by analysis of their ribosomal and mitochondrial DNA. *Ann Trop Med Parasitol* 2007; 101: 621-641.
9. Barragan-Saenz FA, Sanchez-Nava P, Hernandez-Gallegos O, et al. Larval stages of trematodes in gastropods from lake Chicahuapan, State of Mexico. *Parasitol Res* 2009; 105:1163-1167.
10. Dekock KN, Wolmarons CT, Bornman M. Distribution and habitats of the snail *Lymnaea truncatula*, intermediate host of the liver fluke *Fasciola hepatica*, in South Africa. *J S Afr Vet Assoc* 2003; 74: 117-122.
11. Pointier JP, Cazzaniga NJ, Gonzales-Salas C, et al. Anatomical studies of sibling species within neotropical lymnaeids snail, intermediate host of fascioliasis. *Mem Inst Oswaldo Cruz* 2006; 101:431-435.

12. Soliman MFM. Epidemiological review of human and animal fasciolosis in Egypt. *J Infect Dev Countries* 2008; 2: 182-198.
13. Dryffuss G, Abrous M, Vignoles P, et al. *Fasciola hepatica* and *Paramphistomum daubneyi*: Vertical distribution of metacercariae on plants under natural conditions. *Parasitol Res* 2004 ; 94: 70-73.
14. Klockars J, Huffman J, Fried B, et al. Studies on the *Holosoma cercariae* from Douglas Lake, Michigan. *Trans Am Microscopical Soc* 1928; 74: 75-80.
15. Faltynkova A, Haas W. Larval trematodes in freshwater molluscs from the Elbe to Danube rivers (Southeast Germany): Before and today. *Parasitol Res* 2006; 99:572-582.
16. Erasmus DA. *The Biology of Trematodes*. Edward Arnold, London, 1972.
17. Moukrim A, Oviedo JA, Varelle-Morel Ch, et al. *Haplometra cylindracea* (Trematoda: Plagiorchiidae) in *Lymnaea truncatula*: cercarial shedding during single or dual infections with other digenean species. *Res Rev Parasitol* 1993; 53: 57-61.
18. Mansourian A. Fresh water snail fauna of Iran. PhD desertation, Tehran Medical Sciences University, Iran, 1992. (in Persian)
19. Farahnak A, Essalat MA. A study on cercarial dermatitis in Khoozestan province, southwestern Iran. *BMC Public Health* 2003; 3:35-35.
20. Nourpishah SH. The biology of *Lymnaea* snail and its role in transmitting of infection to human and animal in Khoozestan province. MS desertation in medical parasitology, Health Faculty of Tehran Medical Sciences University, Iran, 1998. (in Persian)
21. Zamini GH. Survey of freshwater snails and identification of intermediate hosts of human and animal trematodes in Khoozestan and Kordestan Provinces, Iran. PhD desertation in medical parasitology, Health Faculty of Tehran Medical Sciences University, Iran, 1999. (in Persian)
22. Ashrafi A, Massoud J, Holakuei K, et al. Evidence suggesting that *Fasciola gigantica* may be the most prevalent causal agent of fasciolosis in northern Iran. *Iran J Public Health* 2004; 33: 31-37.
23. Athari A, Gohar-Dehi SH, Rostami M, et al. Determination of definitive and intermediate hosts of cercarial dermatitis-producing agents in Northern Iran. *Arch Iran Med* 2006; 9: 11-15.
24. Gutierrez A, Hernandez DF, Sanchez J. Variation of snail's abundance in two water bodies harboring strains of *Pseudosuccinea columella* resistant and susceptible to *Fasciola hepatica* miracidial infection, in Pinar del Rio Province, Cuba. *Mem Inst Oswaldo Cruz, Rio de Janeiro* 2005; 100(7): 725-727.
25. Faltynkova A, Nasincova V, Kablaskova L. Larval trematodes (digenea) of planorbid snails (Gastropoda: Pulmonata) in Central Europe: a survey of species and key to their identification. *Syst Parasitol* 2008; 69:155-178.
26. Frandsen F, Christensen NO. An introductory guide to the identification of cercariae from African fresh water snails with special reference to cercariae of trematode species of medical and veterinary importance. *Acte Trop* 1984; 41:181-202.
27. Faltynkova A, Niewiadomska K, Santos MJ, et al. *Furcocercous* cercariae (Trematoda) from freshwater snails in Central Finland. *Acta Parasitol* 2007; 52(4): 310-317.
28. Faltynkova A. Larval trematodes (digenea) in molluscs from small water bodies near Ceske Budejovice, Czech Republic. *Acta Parasitol* 2005; 50(1): 49-55.
29. Zajicek D. Cercarie a dalsi vyvojova stadia motolic u plzu z nekterych rybnicnich soustav Jiznich Cech.

- Ceskoslovenska Parasitologie 1963; 10:187-205.
30. Zbikowska E, Kobak J, Zbikowski J, et al. Infestation of *Lymnaea stagnalis* by digenean flukes in the Jeziorak Lake. Parasitol Res 2006; 99:434-439.
 31. Zbikowska E. Infection of snails with birds chistosomes and the threat of swimmers itch in selected Polish lakes. Parasitol Res 2004; 92:30-35.
 32. Ferte H, Depaquit J, Carre S, et al. Presence of *Trichobilharzia azidati* in *Lymnaea stagnalis* and *T. franki* in *Radix auricularia* in Northeastern France: Molecular evidence. Parasitol Res 2005; 95:150-154.
 33. Salahi-Moghaddam A, Mahvi AH, Molavi GH, et al. Survey on *Lymnaea palustris* snail parasitology and its ecological identification in Mazandaran province using of geographical information system (GIS). J Modarres Med Sci 2009; 11(3-4): 65-71.
 34. Horak P, Kolarova L, Adema CM. Biology of the schistosome genus *Trichobilharzia*. Adv Parasitol 2002; 52: 155-233.
 35. Zbikowska E. Thermal behaviour of *Lymnaea stagnalis* (L.) infected with larvae of *Trichobilharzia szidati* (Neuhaus, 1952). Folia Mal 2002; 10:45.