Schistosome cercariae as the causative agent of swimmer's itch in Iceland

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Abstract

During late summer in 1995 to 1997, repeated outbreaks of maculopapular skin eruptions were noted on the legs of children after wading in the pond in the Family Park in Laugardalur, Reykjavík, Iceland. Clinical symptoms developing on the legs resembled those of cercarial dermatitis. An examination of *Lymnaea peregra* snails from this pond and from the adjacent Lake Tjornin resulted in detection of previously undescribed schistosome cercariae. This is the first report of schistosomes in Iceland and also the most northern occurrence of these parasites in Europe.

Introduction

Cercarial dermatitis is a skin allergic response which develops following penetration of larval stages of the trematode family Schistosomatidae. It is diagnosed mainly during the summer months in persons coming in contact with free swimming schistosome cercariae either in fresh water (e.g. genera Schistosoma, Trichobilharzia, Gigantobilharzia) or salt water (e.g. genus Austrobilharzia) bodies (Hutton, 1960; Dönges, 1965; Kolářová et al., 1997). Dermatitis caused by cercariae of non-human schistosomes has been reported from many countries in Asia, North America, Africa, Australia and Europe (e.g. Hoeffler, 1982; Kolářová et al., 1989). In northern Europe, the disease was described in Denmark, Finland and Norway (Pirilä & Wikgren, 1957; Berg & Reiter, 1960; Thune, 1994; Thors et al., 1997). It seems that the European infection agents are represented mainly by cercariae of bird schistosomes (e.g Kolářová et al., 1997), whose intermediate hosts are various species of aquatic snails.

Schistosome cercariae are able to penetrate the skin of various vertebrate hosts. However, their further development

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depends on the compatibility/incompatibility of the infected host. The infection of a compatible host results in the development of adult egg-laying worms. In an abnormal host, however, the parasite dies at various intervals after the penetration. Death of cercariae soon after the penetration was described for example by MacFarlane (1949). Some reports refer to the detection of schistosomula in the lungs of infected hosts (e.g. Haas & Pietsch, 1991; Horák *et al.*, 1998b). Bacha *et al.* (1982) succeeded in finding adult worms of the bird parasite *Austrobilharzia variglandis* in the gerbil (*Meriones unguiculatus*). The possible migration of parasites in an abnormal host underlines the importance of bird schistosomes in medical parasitology.

Clinical symptoms and signs of cercarial dermatitis seem to be dependent on the species involved and on the sensitivity of afflicted persons. Contrary to human schistosomes (*Schistosoma*) the penetration by cercariae of bird schistosomes, such as *Trichobilharzia* and *Austrobilharzia*, results in a more severe reaction. In sensitive persons maculopapular skin erruption accompanied by an intensive itching occurs and occasionally fever, local oedema, enlarged lymphatic nodes and other symptoms may develop (e.g. Kolářová *et al.*, 1994).

During August and early September 1995 and 1996, soon after wading in the pond at the Family Park–Farm

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Yard Zoo in Reykjavík, intensive itching on the skin of some children occurred followed by a rushing eruption. At this time, the reason for the itching remained unknown. By the end of August 1997 there was another outbreak of dermatitis which developed exclusively on the legs which had been immersed in the water. Several hours later, the development of pustules accompanied by oedema was observed. In many cases, the skin reaction was intense, persisted for several days and then disappeared after two weeks. These clinical symptoms resembled those seen in cercarial dermatitis although this disease had not been reported from Iceland.

The aim of the present study was therefore to identify the agent responsible for the dermatitis observed in children who had been wading in the Family Park, Reykjavík, Iceland.

Materials and methods

Characterization of the study sites

The 4000 m² pond in the Family Park in Laugardalur was established in 1993. The most common resident bird species include the greylag goose (*Anser anser*), mallard (*Anas platyrhynchos*), tufted duck (*Aythya fuligula*), and various gull species (*Larus spp.*) which visit the area throughout the year provided the pond is free of ice. In the autumn of 1997 *Lymnaea peregra*, the only snail species occurring in the pond, was present in very high densities.

The other study area from which snails were collected was the adjacent 87,000 m² Lake Tjornin. In the spring of 1997 five duck species (*Anas platyrhynchos, A. strepera, Aythya fuligula, A. marila* and *Somateria mollissima*), the mute swan (*Cygnus cygnus*), greylag goose (*Anser anser*) and arctic tern (*Sterna paradisea*) nested in the vicinity of the lake. The herring gull (*Larus argentatus*), black-headed gull (*L. ridibundus*) and other large gulls are also common. In contrast to the pond in the Family Park, birds occur on the lake in much higher densities. Two snail species *Lymnaea peregra* and *Physa* cf. *integra brevispira* occur in the lake and both are very common.

Parasitological examination

Altogether 162 and six snails of *L. peregra* from both localities and 69 *P.* cf. *integra brevispira* from Lake Tjornin were examined for the presence of schistosome cercariae.

Morphological characterization of cercariae was made on fresh unfixed material (45 specimens) using Nomarski inteference phase contrast and on fixed and mounted cercariae (31 specimens). Cercariae were fixed in hot 4% formaldehyde, stained with borax-carmine and mounted in Canada balsam. Some cercariae were prepared for histological examination; after washing in buffer (20 mM TRIS, 150 mM NaCl, pH 7.8) they were mounted in JB-4 (Polysciences), sections were cut at 3 μ m and stained with Giemsa.

Some cercariae were used for experimental infections using four ducklings (*Anas platyrhynchos* f.dom.), three canaries (*Serinus canaria*) and five mice (SPF ICR *Mus musculus*) were used. The infection dose varied between 300 and 500 cercariae per bird and 100 cercariae per mouse. Birds and mice were infected with cercariae using the methods of Meuleman *et al.* (1984) and Christensen *et al.* (1984) respectively. Faecal samples from the birds were examined for the presence of schistosome eggs from day 7 p.i. Between 14 and 35 days p.i., the birds were killed and their organs (intestine, liver, kidney, lungs, heart, nasal area) and large blood vessels examined microscopically in 0.85% saline for the presence of eggs or parasites. Mice were killed 30 min, 1 h, 4 h, 4 days and 7 days p.i. and their organs (skin, lungs, liver, kidney,

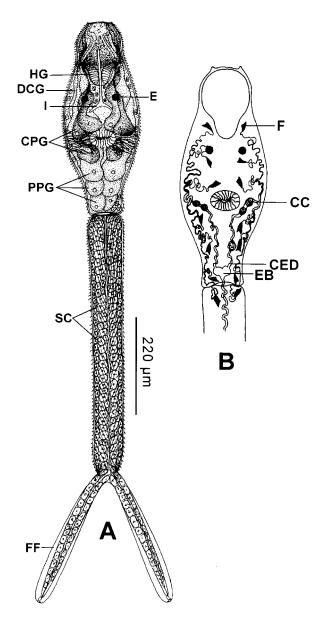


Fig. 1. Schistosome *Cercaria* sp. from Iceland. A. Cercaria. B. Excretory system of cercaria. CC, clusters of cilia; CED, collecting excretory duct; CPG, circumacetabular penetration glands; F, flame cells; FF, finfold; DCG, ducts of glands; E, eyespot; EB, excretory bladder; HG, head organ; I, intestine; PPG, postace-tabular penetration gland; SC, stellar cells.

Table 1. Measurements of the schistosome Cercaria sp. (μm) from Iceland.

	Fresh (min-max)	Fixed (mix-max)
Total length	1361.4 (1180–1406)	1092.9 (965-1165)
Length of body	457.5 (410-490)	285.2 (265-300)
Maximum body width	126.1 (120-135)	88.0 (60-100)
Length of tail stem	580.6 (460-586)	523.5 (450-560)
Maximum width		
of tail stem	86.8 (80-90)	48.7 (35-50)
Furca length	323.3 (310-330)	284.2 (250-305)
Anterior organ length	140.0 (110-180)	101.7 (95-105)
Anterior organ width	120.0 (115-125)	69.3 (68–70)
Acetabulum diameter	40.5 (35-50)	27.5 (23-35)
Eyespot	8.0	8.25
Furca thorns	-	12.0

heart, nasal area) were microscopically examined using native mounts of squashed tissue. If larval stages were found, tissues were prepared for histology using standard techniques, sectioned at $5\,\mu$ m and stained by PAS (McManus, 1946).

Results

Field observation

Snails, identified as *L. peregra* from both localities, released schistosome cercariae: these were found in 13 (7.9%) of 165 snails and 3 (50.0%) of 6 snails examined the Family Park pond and Lake Tjornin, respectively. None of the 65 *P.* cf. *integra brevispira* (Lea 1864) snails from Lake Tjornin were infected.

Larval morphology

Measurements of the cercariae (fig. 1A) are presented in table 1. The body surface and tail stem are covered in fine spines of approximately the same length. The anterior end of the body is occupied by a dominant head organ. In fixed cercariae, the acetabulum lies just behind the centre of the body and two eyespots are located in the anterior third between the head organ and the protrusible acetabulum. The intestine with two short caeca commences subterminally and terminates behind the eyespots. There are two pairs of circumacetabular penetration glands; the second pair surrounds the acetabulum dorsally (fig. 2A), and ends behind the acetabulum.

The distal part of the body behind acetabulum is filled with three pairs of postacetabular penetration glands, the ducts of which are, unlike the circumacetabular gland ducts, very large. Both types of gland ducts pass the cercarial body, through the head organ, terminating at the anterior end of body. Giemsa and borax-carmine staining showed that the contents of the circumacetabular glands remain unstained whereas those of the postacetabular glands are pink-stained (fig. 2B). The excretory system (fig. 1B) is composed of flame cells, clusters of cilia and excretory ducts; two collecting excretory ducts lead into the excretory junction at the base of the body and the main canal continues through the tail system and furcae. There are seven pairs of flame cells and two pairs of cilia

https://doi.org/10.1017/S0022149X00700459 Published online by Cambridge University Press

clusters, which are inside the collecting duct at the level of the acetabulum. The flame cell formula without clusters of cilia is: 2(3+3+(1)) = 14, and with clusters: 2[3+3+(2)+(1)] = 18. The tail is striped and filled by many stellar cells of different sizes, whereas smaller cells lie just under the tegument. The furcae are cross striped and the finfolds are well developed almost along the entire length of the furcae. The furcae terminate by two thorns. The cercariae are photo-sensitive and are able to

Infection experiments

attach to the wall of the container.

Penetration activity on the skin was confirmed by the presence of red maculae on the legs of the two bird species and mice. However, no parasites were detected in the internal organs of the birds, whereas in mice schistosomula were detected in the skin 30 min, 1 h and 4 h p.i. (fig. 3). A few living schistosomula were observed in the epidermal burrows without any sign of a host reaction, although post mortem examinations of mice revealed no parasites in the internal organs.

Deposition of material

Snails

 Lymnaea peregra Laugardalar, Reykjavík, Iceland Deposition of material: Natural History Museum of Vienna, Austria: NHMW Moll.90.677
Physa cf. integra brevispira Lake Tjornin, Iceland Deposition of material: Natural History Museum of Vienna, Austria: NHMW Moll.90.678

Cercariae

Intermediate hosts: Lymnaea peregra

Type locality: Laugardalar and Lake Tjornin, Reykjavík, Iceland

Deposition of material: Natural History Museum of Vienna, Austria: holotype: *Cercaria* sp. Laugardalur, Reykjavík, Iceland NHMW EV 3690; paratype: *Cercaria* sp. Lake Tjornin NHMW EV 3689.

Discussion

The clinical course of the skin disease basically corresponded with the description of cercarial dermatitis in studies by Allerberger *et al.* (1994), Blair & Copeman (1977) and others. Furthermore, an examination of *L. peregra* snails resulted in the finding of schistosome cercariae. Therefore, we assume that repeated outbreaks of cercarial dermatitis in the pond of the Family Park in Revkjavík were caused by the schistosome cercariae.

The identification of the snail shells was confirmed by the Natural History Museum of Prague, Czech Republic (Dr Vaćlav Pfleger), at the Natural History Museum of Vienna, Austria (Dr Helmut Sattmann and Dr Peter Reischütz) and the University of Salzburg (Professor Robert Patzner).

Schistosome cercariae were released from snails of *L. peregra* only. Contrary to known schistosome cercariae of various genera, it is typical for the Icelandic stages that they are of a relatively large size (greater than 1 mm in both fresh and fixed material). Moreover, the comparison of *Cercaria* sp. with cercariae of various genera

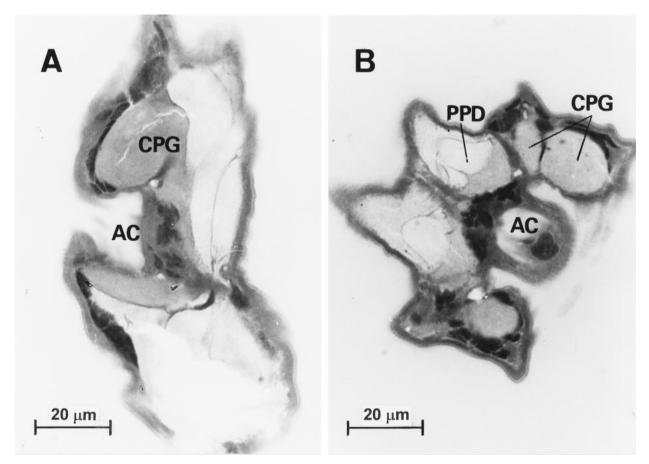


Fig. 2. *Cercaria* sp. transverse sections. A. Second pair of circumacetabular penetration glands (CPG) surrounds dorsally the acetabulum (AC). Section of the cercaria at the level of acetabulum (AC) – three pairs of ducts of postacetabular penetration glands (PPD, unstained) and two pairs of circumacetabular penetration glands (CPG, stained) are visible. (Giemsa staining.)

showed other marked differences: *Austrobilharzia* and *Schistosomatium* possess three pairs of circumacetabular penetration glands, different number of flame cells, no eyespots and no finfolds on the furcae (Penner, 1942; Stunkard & Hinchliffe, 1952) and *Heterobilharzia* cercariae have a different number of flame cells (Trasher, 1964). Furthermore, *Gigantobilharzia* differs in the number of flame cells and in the arrangement of the circumacetabular penetration glands (Dönges, 1965).

The flame cell formula of *Cercaria* sp. agrees with cercariae of *Trichobilharzia*, although the arrangement of *Trichobilharzia* circumacetabular penetration glands differs (*T. szidati*–Kolářová & Horák, 1996; Neuhaus, 1952 and *T. regenti*–Horák *et al.*, 1998a). The same is true for cercaria of the genus *Bilharziella* which differ, moreover, in the ratio of the body/tail lengths (Odening, 1962; Žďárská, 1963). *Cercaria* sp. was found to be similar to *Cercaria ocellata* described in Finland by Wikgren (1956) who recovered schistosome larvae from *Lymnaea stagnalis* and *L. palustris*. However, unlike the cercariae described by Wikgren, the Icelandic *Cercaria* sp. possesses well-developed finfolds along almost the entire length of the furcae as

described in *C. ocellata*. Furthermore, the second pair of circumacetabular glands of *Cercaria* sp. is arranged differently and Wikgren (1956) omitted to mention the presence of two pairs of cilia clusters in the cercariae from Finland.

It is likely that the life cycles of many schistosomes still need to be resolved and that for some known schistosome species only insufficient descriptions of the larval stages are available. Therefore, it is possible that we have characterized a larval stage of a schistosome species whose adult stage might have been previously described.

Considering the definitive host spectrum occurring in the pond of the Family Park in Laugardalur and Lake Tjornin it is possible that we are dealing with the cercaria of *Ornithobilharzia* sp. – adults found in birds of the genera *Larus, Sterna* and others (e.g. Sonin, 1985), *Dendritobilharzia* sp. – adults found in anseriform birds of the genera *Anas, Aythya, Fulica* and *Cygnus* (e.g. Palm, 1965; Price, 1929) or *Trichobilharzia* – adults found in bird species e.g. of *Anas* and *Cygnus* (for review see Azimov, 1975). Furthermore, considering our knowledge of the specificity of the intermediate host, the schistosome genera developing in lymnaeid snails are of special

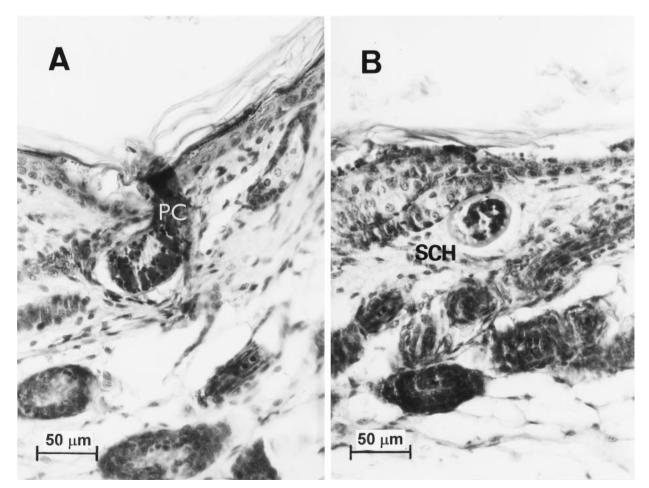


Fig. 3. Experimental infection of mice. A. Living *Cercaria* sp. (PC) penetrating into the dermis directly through the epidermis of a mouse 30 min after exposure; no host reaction is present. B. Living intact schistosomulum (SCH) in the epidermis of a mouse skin; no evidence of host reaction 4 h p.i. (PAS staining.)

interest. Among them, only the cercariae of the genus *Trichobilharzia* are similar morphologically to *Cercaria* sp.

The failure to detect adult parasites in experimentally infected bird hosts may be explained by many factors, for example, inappropriate host species, the course of infection, and insufficient doses of cercariae. The difficulties in identifying the definitive host can be underlined by the fact that mixed populations of various wild freshwater birds, coastal birds and open sea birds exist in Iceland. The detection of schistosomula in mice confirmed the ability of the cercariae to penetrate mammalian skin. The absence of a host reaction, as described by Batten (1956), Haemmerli (1953) and others was probably due to the short time interval of infection during which the experimental procedures were performed.

In conclusion, it was demonstrated for the first time that schistosomes occur in Iceland and that there exists a risk of infection of cercarial dermatitis. Moreover, Iceland represents the most northern occurrence of schistosome cercariae in Europe.

Acknowledgements

The investigation was partially supported by the grant of the Ministry of Public Health of the Czech Republic (IGA MZ CR No. 4945-3). The authors are grateful to Thorbjorg Kristjansdottir and Sigrun Thorlacius, Family Park in Reykjavík who assisted with the collection and examination of the snails, to Dr Jens Magnusson at the Medical Health Centre in Grafarvogur, Reykjavík who provided the information on the clinical symptoms of the affected children. The authors are also indebted to Austrian specialist Dr Helmut Sattmann from Natural History Museum of Vienna, Dr Peter Reischütz Gymnasium, Horn and Professor Robert Patzner, University of Salzburg and Dr Václav Pfleger from Natural History Museum, Prague, Czech Republic for their valuable discussion about the snail determination. Finally, Jan Dvořák, a student in the Department of Parasitology, Faculty of Science, Charles University, Prague, Czech Republic is acknowledged for technical assistance.

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(Accepted 6 January 1999) © CAB International, 1999