

Research Note

Results on search for the broad fish tapeworm *Dibothriocephalus latus* (Linnaeus, 1758), (syn. *Diphyllbothrium latum*) (Cestoda: Diphyllbothriidea), in the Danube River

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Summary

Diphyllbothriosis is a fish-borne parasitic zoonosis caused by so-called “broad tapeworms” or “fish tapeworms” of different genera of the order Diphyllbothriidea. *Dibothriocephalus latus* (Linnaeus 1758), (syn. *Diphyllbothrium latum*), is a medically important type species of the genus, whose occurrence in various European regions is either regular, e.g. in the Alpine lakes region, or occasional and sporadic, e.g. in the Danube River region. For the latter, data on the detection of *D. latus* plerocercoids in the second intermediate fish host (European perch *Perca fluviatilis*), as well as in definitive hosts (human and dog), in which infection was directly linked to the consumption of infected fish from the Danube, were published more than 50 years ago. In order to assess the current situation, we aimed to find out whether *D. latus* is present in the natural environment of the Danube River. In total, 700 perch from five sampling sites in the Slovak part of the Danube River were examined. Plerocercoids were not detected in any fish examined, which leads to the conclusion that *D. latus* is currently not present in the studied aquatic environment.

Keywords: European perch; plerocercoid; diphyllbothriosis; fish-borne zoonosis

Introduction

Diphyllbothriosis, caused by so-called “broad tapeworms” or “fish tapeworms” of different genera of the order Diphyllbothriidea, is a significant fish-borne parasitic zoonosis responsible for about 20 million human infections worldwide (Chai *et al.*, 2005). The type species of the recently resurrected genus *Dibothriocephalus* is the medically important tapeworm *Dibothriocephalus latus* (Linnaeus 1758), (syn. *Diphyllbothrium latum*) (Waeschenbach *et al.*, 2017). It utilizes copepods as the first intermediate hosts and freshwater fish (in Europe mainly European perch *Perca fluviatilis*, Northern pike *Esox lucius* and burbot *Lota lota*) as the second intermediate hosts. Humans and carnivorous mammals serve as definitive hosts. Humans can be infected with *D. latus* plerocercoids by eating raw or undercooked fish. The infection can be either au-

tochthonous (the infected fish originated from local rivers or lakes) or imported (the infection was acquired by consumption of fish during a stay abroad). The Food and Agricultural Organization (FAO) of the United Nations and the World Health Organization (WHO) included diphyllbothriosis on the list of 24 medically and veterinary important fish-borne parasites (Robertson *et al.*, 2013). Understandably, knowledge on the occurrence and geographic distribution of *D. latus*, its circulation in the natural environment, and determination of possible risks of infection are crucial. Based on the frequency of occurrence of *D. latus* in Europe, there are regions with frequent or relatively frequent occurrence, areas with sporadic or imported cases and countries where diphyllbothriosis has not been reported. Frequent occurrence of the parasite in humans, as well as in the natural environment, has been recorded e.g. in the Alpine lakes region, including Switzerland,

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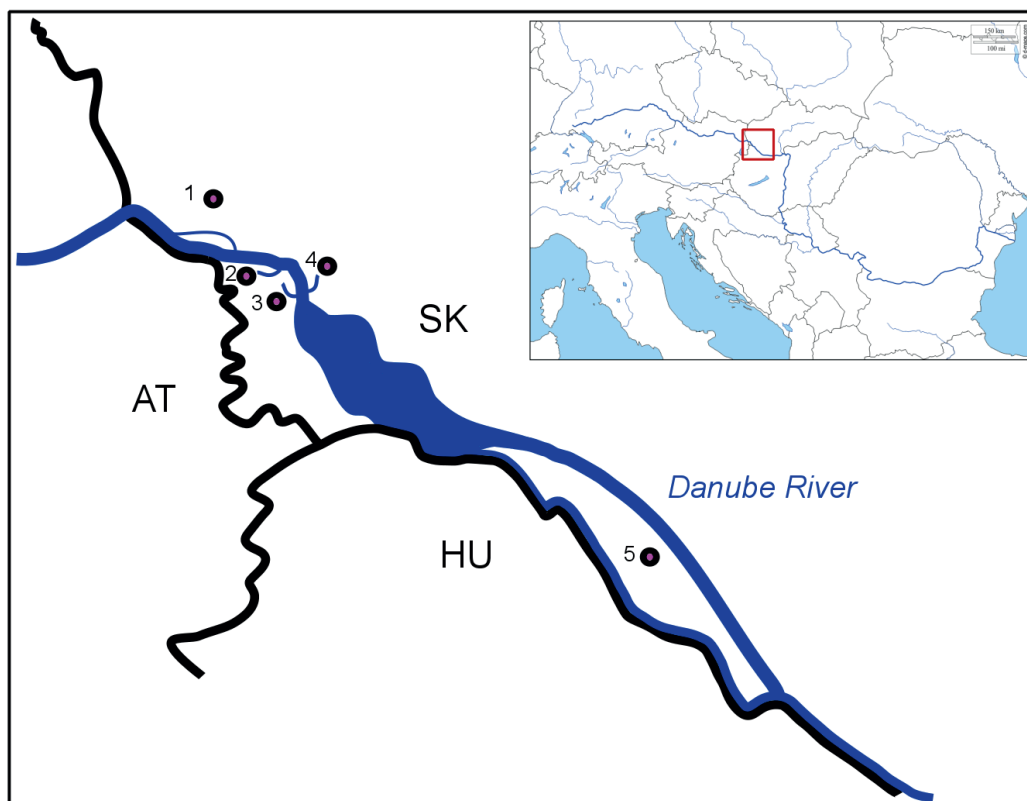


Fig. 1. Schematic representation of the entire flow of the Danube River (right upper corner) and graphical scheme of sampling sites; (1) Karloveské river arm; (2) Starohájske river arm; (3) Jarovecké river arm; (4) Biskupické river arm; (5) pond Bodíky-materiálová jama B. Abbreviations: SK, Slovakia; HU, Hungary; AT, Austria.

eastern France and northern Italy (Yera *et al.*, 2008; Wicht *et al.*, 2010; Dupoy-Camet, 2015; Gustinelli *et al.*, 2016). Regions with sporadic occurrence are localities with imported human cases, e.g. Czech Republic (Vlková *et al.*, 2007) and Romania (Stanciu *et al.*, 2009), and regions with the occasional findings of the tapeworm in fish, e.g. Ireland (Harris & Hickey, 1945), Serbia (Petrovic, 1972 cited in Djikanovic *et al.*, 2012) and the Danube River (Roman, 1955).

The Danube River plays a notable role in spreading various parasitic and infectious diseases of fish, since it is shared by 10 European countries (Germany, Austria, Slovakia, Hungary, Croatia, Serbia, Romania, Bulgaria, Moldova and Ukraine), making it the most international river basin in the world (Liska, 2015). According to several studies, the middle Danube (the part of the river between Slovakia and Serbia/Romania) certainly represents a great risk for native fish communities due to the presence of different pathogenic parasites, such as the nematode *Anguillicola crassus* or the rosette agent *Sphaerothecum destruens* (Gozlan *et al.*, 2005, 2009). Concerning diphylobothriosis, only a few published papers have addressed either a direct presence of larval stages (plerocercoids) of *D. latus* in fish from the Danube River (Roman 1955) or documented autochthonous *D. latus* infections linked with the consumption of raw perch from the Danube in humans (Čatár *et al.*, 1967) and dogs (Supperer & Wenzel, 1967). These data were

published more than 50 years ago and can be classified as sporadic. The presence of *D. latus* in the Slovak part of the Danube River was mentioned in other papers dealing with summary data on tapeworms in Slovakia (Macko *et al.*, 1993, Moravec, 2001) and a review of fish parasites in Slovakia (Žitňan, 1982). However, these publications referred to a single human case report published by Čatár *et al.* (1967), and examinations of fish have never been performed.

Several other papers have been published on *D. latus* occurrence in the Danube countries (e.g. Serbia, Romania and Moldova). However, majority of those data were either not based on direct observations of the authors, or proper taxonomic identification, details on location of plerocercoid in fish and geographic locality were missing. Plerocercoids of *D. latus* were found in pikes in the Danube Delta district called Mila in Romania and from the Prut River (tributary to the Danube River) in Moldova (von Bondsdorff, 1977). Djikanovic *et al.* (2012) listed occurrence of *D. latus* in perch and salmonid *Salmo letnica* from Serbia (data originally published as a conference abstract by Petrovic, 1972). However, precise geographic locality was not provided, therefore, this finding cannot be reliably related to the Danube River. Another paper dealing with *D. latus* in perch in Romania was published by Goga *et al.* (2014); based on the figures of the tapeworm, this finding seems to be a misidentification.

The aim of the current work was to find out whether *D. latus* is present in the natural environment of the Danube River in order to assess the current situation. Since relevant data were published more than a half-century ago, and due to medical importance of diphyllobothriosis, up-to-date data on the occurrence of *D. latus* in the Danube River are needed.

Material and Methods

Parasitological examinations of 700 European perch from four arms of the Slovak part of the Danube River and one artificial pond filled with water from the Danube (Fig. 1) were carried out in autumn 2017 (October) and spring-summer 2018 (April and June). In particular, 86 fish originated from the Karloveské river arm (48° 8'46.08"N, 17° 3'50.33"E); 314 perch were examined from the Starohájske river arm (48° 6'11.50"N, 17° 7'56.19"E); 119 fish came from the Jarovecké river arm (48° 4'32.34"N, 17° 8'23.90"E); 98 were examined from the Biskupické river arm (48° 5'15.45"N, 17° 9'44.21"E); and 83 perch were caught in the pond called Bodíky-materiálová jama B (47°56'26.66"N, 17°25'42.55"E). Fish were provided to us under a commercial service by certified fishermen. Considering that a significant majority of plerocercoids collected from perch have been found in the fillet (Gustinelli *et al.*, 2016), all fish were examined by detailed filleting of the entire musculature. In addition, the peritoneal cavity, intestine, liver and other abdominal organs were also checked.

Ethical Approval and/or Informed Consent

The research related to animals has been complied with all the relevant national regulations and institutional policies for the care and use of animals.

Results and Discussion

As the result of our examinations, no *D. latus* plerocercoids were found in any of the 700 fish from five studied localities of the Danube River in Slovakia. Instead, the nematode *Eustrongylides* spp. and the fluke *Clinostomum complanatum* were found in the musculature. In addition, the tapeworm *Proteocephalus percae* was detected in the perch intestine and larvae of the tapeworm *Triaenophorus nodulosus* were isolated from the liver cysts (details not provided). In general, determination of plerocercoids in fish or adult tapeworms in definitive hosts provides direct evidence on the occurrence of *D. latus* in the local environment. The oldest data on the presence of *D. latus* were published by Roman (1955); the author listed the detection of a single *D. latus* plerocercoid from the intestine of a European perch from the Danube Delta in Romania. However, the information provided by Roman (1955) is very brief, lacking details on morphological description or any other more species-specific data. Moreover, *D. latus* is preferably localized in the upper mass of the musculature of the European perch, espe-

cially in the dorsal muscles (Gustinelli *et al.*, 2016).

Later, Supperer and Wenzel (1967) reported on a dog fed with fish from the Danube River being infected with *D. latus*. In the same year, a detailed and well documented human case report was provided by Čatár *et al.* (1967). A local fisherman, who declared regular eating of raw perch caught in the Slovak part of the Danube River, indicated gastro-intestinal problems. After medical examination, diagnosis and relevant anthelmintic treatment, a 7.5 m long *D. latus* tapeworm was released from the patient. Thanks to the detailed case report and epidemiological anamnesis of this particular human infection, the source of infection was detected, and it was overwhelmingly linked with the patient's habit of consuming raw perch caught in the Danube River.

Taking into consideration a medical importance and zoonotic potential of *D. latus*, it is rather incomprehensible, why no recent reliable data on its occurrence in fish from the Danube River have been published. Did the infection persist in this local environment but somehow manage to escape the attention of local ichthyologists and parasitologists? Or, was the prevalence of the infection so low that diphyllobothriosis was overlooked and not recorded? Finally, what was the reason for possible disappearance of *D. latus* from the Danube River?

Concerning the ecological and biological conditions crucial for maintaining the life cycle of *D. latus*, apart from a presence of a dominant second intermediate fish host (European perch) also the first intermediate host (Copepoda) (Vadadi-Fülöp, 2009) is present in the Danube River. However, *D. latus* plerocercoids have been more frequently found in fish coming from backwater, mainly lakes e.g. Alpine lakes of northern Italy (Gustinelli *et al.*, 2016), Switzerland (Wicht *et al.*, 2010), eastern France (Yera *et al.*, 2008) and rarely in fish originating from rivers, e.g. the Kokemäenjoki River in Finland (Wikgren, 1963). The explanation for why some Alpine lakes represent localities with ongoing detection of *D. latus* plerocercoids in perch and adult tapeworms in humans is the ineffective sewage treatment systems leading to the contamination of *D. latus* eggs shed by infected humans, and possibly also other mammals. In addition, consumption of a local raw fish delicacy called carpaccio results in the persistence of human *D. latus* cases in the Alpine lakes region (Gustinelli *et al.*, 2016). It is evident that human play important role in maintaining the life cycle of *D. latus* in the Alpine region. On the contrary, the Danube River represents a more dynamic biotope with possible barriers for the maintenance of the life cycle stages of *D. latus*. The absence of *D. latus* in the Danube River is also supported by latest preliminary data on dissection of 32 European perch from the Danube Delta in Romania, where no *D. latus* plerocercoids have been found (M. Oros and D. Barčák, Institute of Parasitology, SAS, personal communication).

In conclusion, the localities with old findings of *D. latus*, sporadic detection of the parasite, imported human cases and random determination in the natural environment, had to be considered carefully and deserve further attention for revealing the actual epidemiological and ecological situation. Based on the amount of

fish investigated in our study, *D. latus* is very probably not present in the middle part of the Danube River. Novel data on ichthyoparasitological surveys, further examinations and epidemiological studies are needed in order to create the latest distribution map of *D. latus* in Europe.

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Conflict of interest

Authors declare no conflict of interest.

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