### Troglotrema acutum (Digenea) from carnivores in the Czech Republic

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### Summary

A collection of 5 282 skulls of 14 carnivore species from the Czech Republic was examined and characteristic changes of the visceral part of cranial skeleton caused by Troglotrema acutum were found in 6 of the studied species: Mustela putorius (prevalence 7.1 %), Martes martes (1.6%), Martes foina (0.6%), Meles meles (1.1%), Lutra lutra (0.9 %) and Vulpes vulpes (0.8 %). The typical host of this trematode species is *M. putorius*; other carnivore species are accidental hosts. Fresh material (225 carnivores of 7 species) was examined for accurate identification of cranial skeleton changes and determination of parasite species. Nine adult trematodes from 3 infected M. putorius were obtained. T. acutum has been found on 8.1 % of the territory of the Czech Republic so far. Most often it was detected at localities at altitudes from 201 to 300 m a.s.l. (55.3 %). Its distribution in the Czech Republic is of focal character. It was found that neither spatial nor vertical distributions of T. acutum correspond to the present distribution of the first intermediate hosts determined so far (small prosobranchiate snails of genus Bythinella). A hypothesis has been formulated that the first intermediate hosts of this trematode species may also be other species of prosobranchiate snails, whose distribution is not limited by altitude.

Key words: *Troglotrema acutum*; polecat; distribution; ecology

### Introduction

*Troglotrema acutum* (Leuckart, 1842) and some nematode species from the genus *Skrjabingylus* Petrov, 1927 are found in the nasal sinuses of carnivores (mainly mustelids). These parasites inflict serious cranial lesions in their hosts. Some authors, Baer (1931), Haltenroth (1938), Kontrimavichus (1969) and Staněk (1961, 1963) suggest that it is possible to morphologically distinguish the lesions caused by the trematode from those caused by nematodes.

The geographical distribution of *T. acutum* is notable, it covers only the countries of central, southern and western Europe (Skrjabin 1949, Yamaguti 1971). The first finding of *T. acutum* in the territory of the Czech Republic was reported by Ullrich (1930), the possibility of more frequent occurrence was brought up by Dyk (1960) and factual findings were described by Staněk (1961,1963), Svatoš (1963), Ašmera (1985, 1986) and Tenora and Staněk (1989). Hanák and Beneš (2001) studied the significance of regional geographical distribution (Northern Moravia, Czech Republic) of nasal parasites in carnivores; however, they did not differentiate the lesions caused by the trematode (*T. acutum*) and nematodes (*Skrjabingylus*) species.

From the number of the findings published so far and from their time distribution and localities of occurrence it is possible to conclude that *T. acutum* is a relatively rare parasite in the Czech Republic as well as in its whole home range. While the collection of fresh material of mustelids for parasitic examination in sufficient amount is complicated, investigation of museum skulls and identification of specific cranial lesions is possible and effective.

This paper presents the results of the study of prevalence and distribution of *T. acutum* performed with abundant carnivore skull material from various museum collections and fresh material from the Czech Republic. The number of localities covers a substantial part of the country's territory, which enables us to assess the distribution of this parasite quite accurately and to analyse the species spectrum of its final hosts in the studied space and time. Some ecological relationships determining the infection of the main host *M. putorius* (intermediate hosts, altitude, focal character of infections) and other aspects of this parasitehost system are analysed.

### **Material and Methods**

The presence and morphology of lesions caused by trema-

Species	NM Prague		MM Brno		SM Opava		IVB CzAS		Total	
1	n	Positive	n	Positive	n	Positive	n	Positive	n	Positive
Mustelidae										
M.nivalis	141	0	68	0	699	0	346	0	1254	0
M. erminea	59	0	64	0	99	0	102	0	324	0
M. putorius	189	2	232	20	189	38	547	22	1157	82
M. eversmannii	21	0	29	0	5	0	14	0	69	0
M. martes	67	1	128	1	72	2	175	3	442	7
M. foina	139	1	153	2	177	0	243	1	712	4
M. vison	14	0	18	0			24	0	56	0
L. lutra	86	0	11	0	4	0	10	1	111	1
M. meles	100	0	37	0	16	0	31	2	184	2
Felidae										
L. lynx							75*	0	75	0
F. silvestris							1	0	1	0
Canidae										
V. vulpes	189	2	246	2	96	1	347	0	878	7
N.procyonoides	2	0		_	7	0	10	0	19	0
Procyonidae										
P. lotor							2	0	2	0
Total	1007	6	986	25	1364	41	1927	35	5282	101

Table 1. Survey of the examined species of carnivores in the Czech Republic

NM Prague – National Museum Prague; MM Brno – Moravian Museum Brno; SM Opava – Silesian Museum, Opava; IVB, CzAS – Institute of Vertebrate Biology CzAS, Brno; \* – Červený and Koubek (2000)

todes in 5 282 skulls of carnivores (11 autochthonous species and 3 allochthonous species) from Czech Republic were evaluated. The studied skulls were collected over a relatively long period (1949-2002) and were obtained from four skull collections in the Czech scientific institutions: National Museum, Prague, Moravian Museum, Brno, Silesian Museum, Opava and Institute of Vertebrate Biology CzAS, Brno. This skull material was prepared in a standard way as dried museum material. Apart from the dry material, we also examined 39 fresh skulls of Mustela putorius, 12 skulls of M. eversmannii, 3 skulls of M. erminea, 5 skulls of M. nivalis, 24 skulls of M. vison, 62 skulls of Martes foina, 5 skulls of M. martes, 74 skulls of Vulpes vulpes and 1 skull of Nyctereutes procyonoides. From 3 infected specimens of M. putorius we obtained 9 adult T. acutum (Fig. 1). The numbers of carnivores examined from individual collections are presented in Table 1.

Skulls were regarded as damaged by the *T. acutum* infection when they did not show lesions caused by preparation, shots or strokes and when they exhibited some of the following changes in the parasitised cranial skeleton: 1. Evident asymmetry; 2. Perforations, even in the absence of asymmetry; 3. Presence of considerably large thinned areas of the bone wall when the cranium was otherwise robust. A typical attribute of cranial lesions caused by *T. acutum* infection is the changed bone structure over a large area, which may cover the whole nasal part of the skull. The bone perforations are large and notably asymmetric with



Fig. 1. Troglotrema acutum (Leuckart, 1842) from Mustela putorius (L.). Total view (Original)

uneven edges (Fig. 2). In each of the findings we recorded the type of the defect and number of perforations. To express the geographical distribution of *T. acutum* we used solely the material with exactly determined locality of origin and date of collection. Standard grid maps were used for the mapping the distribution of *T. acutum*. This system is based on geographic coordinates (P6' x 10') and the territory of the Czech Republic is divided into quadrate format tetragons of approximately 11.2 x 12 km. A similar approach has been applied in the mapping of mammals and birds (Anděra and Hanzal, 1996).



Fig. 2. Polecat skull (*Mustela putorius*) with changes typical for *Troglotrema acutum* trematode infection (Photo by R. Blažek)

#### Results

# 1. Species spectrum of hosts and prevalence values of T. acutum

Out of the total number of 14 carnivore species of 4 families living in the Czech Republic (Table 1), the infection with *T. acutum* was found only in hosts from Mustelidae (5 species) and Canidae (1 species). The highest prevalence value (in %) was found in *M. putorius* (7.1). Lower values were found for *Martes martes* (1.6), *Meles meles* (1.1), *Lutra lutra* (0.9), *Martes foina* (0.6) and *Vulpes vulpes* (0.8). The results suggest that *M. putorius* is the main host of *T. acutum*.

## 2. Frequency of occurrence of perforations and individual types of damage to the visceral part of cranial skeleton

A detailed analysis of this parameter could be performed only for *M. putorius* due to the sufficient number of positive individuals (Fig. 3). In this host the number of perforations found in the visceral part of cranial skeleton was within a wide range, from 1 to 13. Out of the affected skulls, 34.2 % had only one perforation, 17.1 % two, 14.6 % three, 9.8 % four and 7.3 % five perforations. Higher numbers of perforations from 6 to 13 have very low frequency (3.7 – 1.2 %). In the rest of the hosts the infection intensity was in most cases relatively low. In *M. martes* only 1 - 3 perforations were found, in *M. foina* 1 - 4 and in *V. vulpes* we found 1 - 2 perforations.

Total and often multiple perforations of bones in the nasal part of the skull were detected in most of the hosts. All of the positive cases combined 2 or 3 types of skeleton damage. Only in the case of *L. lutra* and *M. meles* did the infection manifest itself by the formation of net-shaped deformation of frontal and part of the parietal bones. The data on frequency of the individual types of skull damage are presented in Table 2.



Fig. 3. Quantification of damage to the visceral part of the cranial skeleton in *M. putorius* according to number of perforations

Table 2. Frequency (%	) of the occurrence	e of different type	s of cranial
skeleton lesions i	n carnivores caused	l by T. acutum inf	rection

Species	Positive	Net	Asymmetry	Perforation
		thinned		
Mustela putorius	82	1	1	82
Martes martes	7	1		7
Martes foina	4			4
Lutra lutra	1	1		1
Meles meles	2	2		
Vulpes vulpes	7	1		7

## 3. Geographical distribution of T. acutum in the Czech Republic

The occurrence of *T. acutum* has been confirmed on 35 map squares, that is 5.6 % of the total number of 628 squares covering the territory of the CzR (Fig. 4). The dominant position of *M. putorius* in the natural range of *T. acutum* is proved by the fact that the localities with positive findings in *M. putorius* alone or with other carnivores cover 33 map squares (i.e. 94.3 % of the infected area of the CzR). Positive findings in hosts (*M. martes, M. foina, M. meles, Lutra lutra*) other than *M. putorius* were detected in

6 squares (15.4 %) only. If we complement the positive findings of *T. acutum* from our material with previously published data, then this trematode has been reported from 51 squares so far, which represents 8.1 % of the area of the CzR. Comparison of the localities with *T. acutum* with the mapping grid indicates the presence of preferred smaller or larger regions bound to river or pond ecosystems in southern Bohemia, Českomoravská vrchovina Highlands and especially in northern and southern Moravia.



Fig. 4. Geographical distribution of *T. acutum* in the Czech Republic (1 – our findings; j – other published data)

4. Vertical distribution of *T. acutum in the Czech Republic* Vertical distribution of *T. acutum* was recorded for all findings of infected hosts. Positive findings appeared in hosts from localities with altitudes from 167 to 745 m a.s.l. The majority of the findings (91.1 %) lay under 450 m a.s.l., and 55.3 % of the findings were from the range 201-300 m a.s.l. (Fig. 5).



Fig. 5. Vertical distribution of *T. acutum* findings in the Czech Republic

### 5. Focal character of T. acutum infections

Geographical aspects of *T. acutum* distribution indicate the existence of foci of this parasite. We consider the repeated

findings of this parasite at the same locality (same map square) and within the period of two and more years to be the evidence of the existence of these focuses. Repeated findings of T. acutum in M. putorius appeared in 14 squares in the period from 1957 to 2001 (Table 3). M. putorius was the single host at 64.3 % of the infected area (9 squares). Common infection with M. martes was found at 21.4 % of the area (3 squares) and with V. vulpes at 14.3 % of the area (2 squares). The significance of *M. putorius* as the main host for persistence of the focuses of T. acutum infection is also confirmed by the fact that in other carnivores the infection with this trematode was found at 7 squares only, but always in a single host species and in a single year (in V. vulpes - 3 localities, M. martes, L. lutra and M. meles - 1 locality). Only in one case at one locality was the infection detected in two host species (M. martes, M. foina).

 

 Table 3. Localities of repeated occurrence of T. acutum in the Czech Republic

Quadrat	Locality	Repeated	Time	Host (s)*
Nos.		findings	range	
6167	Zábřeh n. Moravě	7	57 – 59	<i>M.p., M.ma.</i>
6962	Rouchovany	3	89 - 01	М.р.
6965	Židlochovice	5	62 - 87	М.р., М.та.
7065	Pohořelice	2	69 – 84	M.p., V.v.
6865	Bosonohy	4	62 - 73	M.p. M.me.
6072	Hlavnice	10	67 - 71	М.р., М.та.
6073	Opava	3	64 - 66	<i>M.p.</i> , <i>V.v</i> .
6275	Polanka	2	61 - 71	<i>M.p.</i>
6172	Štáblovice	5	62 - 74	<i>M.p.</i>
6174	Háj	4	67 - 70	<i>M.p.</i>
6074	Kravaře	13	58 - 91	<u>М</u> .р.

\*) M.p. – Mustela putorius; M.ma. – Martes martes; M. me. – Meles meles; V.v. – Vulpes vulpes

### Discussion

*T. acutum* was originally described by Leuckart (1842) as *Distoma acutum*. Host of the type material was *M. putorius* from Germany. This trematode was later classed into a newly created genus, *Troglotrema*, by Odhner (1914), who revised the collection of trematodes obtained by Moniez (1890) from polecats in France. Odhner's description together with the description and illustration of an adult trematode by Baer (1931) present the most detailed data about morphometry of this species so far. The trematodes obtained from our material fully correspond to the descriptions of *T. acutum* published up to the present.

Unfortunately, most of the papers about the occurrence and distribution of *T. acutum* in the Czech Republic as well as in other regions published so far are based on the presence of changes in the visceral part of the skull (Staněk, 1961; Schmidt, 1993; Hanák and Beneš, 2001). Although some authors express their doubts (Olt, 1929; Kotlán, 1960), such changes may also be caused by nematodes of the ge-

nus *Skrjabingylus* (Hansson, 1968; van Soest *et al.*, 1972). Thanks to the studies performed on numerous collections of sculls deformed by *T. acutum* and by nematodes of the genus *Skrjabingylus*, the morphological differences in the shape as well as size of the cranial bone perforations caused by these parasites have been described (Baer, 1931; Kontrimavichus, 1969). Still, the determination of the causal agent of these changes is often very difficult and inaccurate (Haltenroth, 1937; Hansson, 1968; Schmidt, 1993; Hanák and Beneš, 2001). This is true especially for the findings of cranial bone perforations and other similar changes in accidental hosts – small carnivores, where the prevalence is extremely low. The only unambiguous evidence in such cases should be the examination of fresh material and finding of the parasite.

The results published so far show that the dominant host species of T. acutum is M. putorius (Haltenroth, 1937; Marconcini and Tasselli, 1969; Artois et al., 1982). Other species of mustelids (M. martes, M. foina, M. meles, L. *lutra*) and canids (V. vulpes) may be without exceptions classified as accidental hosts of this trematode species (Ašmera, 1986; Hanák and Beneš, 2001). However, its occurrence in M. nivalis and M. erminea is not quite clear. T. acutum was not found in M. nivalis and M. erminea until 1972 and Hansson (1968) and van Soest et al. (1972) exclude the possibility of its occurrence in these two species. The finding of T. acutum in M. nivalis reported by Haltenroth (1937) was not confirmed by the study of the same skull material performed by Hansson (1968) and therefore it is improbable. Equally improbable are reports about the occurrence of T. acutum in steppe species such as Vormela peregusna (Haltenroth, 1937) and Mustela eversmannii (Tenora and Staněk, 1989), or in an unspecified seal species (Haltenroth, 1937).

As for the farm animals, Sprehn (1955) found *T. acutum* in *Mustela vison, Mustela putorius* f. *furo* and *Vulpes vulpes fulva*. On the basis of these results he predicted the occurrence in free-living *V. vulpes* too. The possibility of *T. acu-tum* infection in exotic species of carnivores is shown by the skull of *Nasua nasua* (Procyonidae), which shows typical perforations; this animal came from the Zoological garden in Prague (the skull is deposited in the National Museum in Prague).

A drawback of the skull material research lies in the impossibility of evaluating the intensity of the infection. The results of autopsies of fresh material by Artois *et al.* (1982) show quite clearly that it is absolutely impossible to deduce the infection intensity from the number of perforations.

It is thought that the natural range of *T. acutum* is limited to Central Europe (Skryabin, 1949; Kontrimavichus, 1969; van Soest *et al.*, 1972). This fact is documented by the location of most of the published findings from Germany (Pohl, 1912; Förster, 1914; Olt, 1929; Stroh, 1929; Sprehn, 1955; Lehmensick, 1942; Dialer, 1938; Vogel and Voelker, 1978); France (Moniez, 1938; Artois *et al.*, 1982), Austria (Schumacher, 1929), Switzerland (Baer, 1931, 1932; Wegelin, 1930), Italy (Marconcini and Tasselli, 1964), the

Czech Republic (Ullrich, 1930; Staněk, 1961, 1963; Vojtková and Křivanec, 1970; Ašmera, 1985, 1986; Hanák and Beneš, 2001, Faltýnková et al., 2001), Slovakia (Mituch, 1972; Rajský and Porubčanský, 1989) and Poland (Grabda-Kazubska, 1977). The data about the occurrence of T. acutum published by Haltenroth (1937) seems highly improbable. Although it is possible that some of his localizations (Sweden, Romania, Bulgaria or Kaliningrad region) may prove to be correct in the future, localizations of T. acutum in Africa, Minor Asia and Venezuela are absolutely implausible. According to the present knowledge of the range of T. acutum we can say that the eastern margin of its documented occurrence is in Poland and Slovakia. There is no doubt about the absence of T. acutum in the European part of former USSR (Ukraine, Belarus, Lithuania) as none of the many papers on helminthofauna of carnivores brought positive results (Skrjabin, 1949; Kontrimavichus, 1969; Kozlov, 1977; Merkuscheva and Bobkova, 1981; Anisimova, 2002). From the viewpoint of zoogeography it is a very interesting feature that does not correspond to the distribution of the final host.

The distribution of T. acutum is not continuous, even within its natural range. It concentrates in foci, which appear and remain for extended period in biocoenoses suitable for the completion of the life cycle of this trematode. The life cycle of T. acutum was studied in detail by Vogel and Voelker (1978). Experimentally the larval stages up to the cercaria developed in small prosobranchiate snail from genus Bythinella (B. dunkeri). The second intermediate host in which the metacercariae encysted was the frog Rana temporaria. Vojtková and Křivanec (1970) and Grabda-Kazubska (1977) confirmed the presence of metacercariae in free-living frogs R. temporaria, Bombina variegata and Bufo bufo. However, the distribution of T. acutum indicates that the first hosts may also be other species of Bythinella. This prediction was note to Artois et al. (1982) who expect B. compressa and B. austriaca to be the intermediate hosts. Probable occurrence of cercaria T. acutum in B. austriaca in Czech Republic, Poland and Slovakia was also reported by Faltýnková et al. (2001) and Faltýnková and Literák (2002). These presumptions about the wider range of snail species that can act as first intermediate hosts are confirmed by our conclusion that the infection of the typical host of T. acutum (M. putorius) is not geographically and sea level limited only to cool well waters, as assumed by Vogel and Voelker (1978). Most of the infections of M. putorius with T. acutum that we detected were found in lower localities with higher presence of stagnant waters, which contradicts the opinion of Vogel and Voelker (1978) that "diese Schenckengattung und der Parasit sind beide an Berg- und Hügellander gebunden, fehlen degemäss in den Tiefebenen." They also say: "In Böhmen fehlen Bythinellen in den westlichen, nördlichen und südlichen Gebieten, kommen jedoch im östlichen Teil vor, gerade dort, wo der einziege Troglotrema- Herd liegt, der aus der Tschechoslowkei bekannt ist."

Our results describing the distribution and prevalence of *T*.

acutum in M. putorius and some other carnivores lead us to predict that the range of intermediate snails is probably even wider and includes also some other species of prosobranchiate snails. Unlike the species of Bythinella, their which distribution and occurrence in the Czech Republic is neither geographically not sea level strictly limited (Beran, 1998). The infection focuses and their existence in time and space, as well as the significance of the typical final host result from the biological relations in the food chain of the particular biocoenosis. It is known that polecats have either movements only in a restricted range or more or less linear movements within the home range (Lodé, 1999). The capture of anurans is easy for polecats (Weber 1989), and frog aggregation at spawning sites in spring (Blab, 1986; Guyétant, 1986) probably facilitates predation.

Studies on polecat ecology in Europe indicate that amphibians form a significant part of its diet throughout the year. During the vegetation season it is 17.6 - 84.8 % (V) and in winter months still 17.6 - 40.7 % (Wolsan, 1993). In the diets of other mustelids amphibians are represented far less, which corresponds to the prevalence of *T. acutum* in these species. However, if their home ranges overlap or encroach upon the focus of *T. acutum* infection, these species can also take part in the infection cycle.

It is possible to interpret our results as follows: occurrence of *T. acutum* on the territory of the Czech Republic is only sporadic and the active infection focuses are mainly in the localities with lower altitude. Besides the headstream regions it can be also found at localities with prevalence of stagnant water reservoirs. Due to the high pathogenicity of *T. acutum* (see Olt, 1929; Baer, 1931; Dialer, 1938; Artois *et al.*, 1982), this helminthosis may be an important factor influencing the population density of polecat in the Czech Republic. Nevertheless, it is not possible to confirm a direct relationship between the decrease of population density of polecat in the Czech Republic and prevalence of *T. acutum* infection during the last 10 years.

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