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Short communication

Mass leech infestation of sculpin fish in Lake Baikal, with clarification of disease-prone species and parasite taxonomy

Irina A. Kaygorodova *, Natalya V. Sorokovikova

Limnological Institute, 3 Ulan-Batorskaya str., Irkutsk 664033, Russia

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ABSTRACT

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Keywords: Piscine parasite Baicalobdella Baikal sculpin fish Leeches Fish disease Sculpin fish bdellosis in Lake Baikal is caused by leech ectoparasites, which are identified as belonging to the genus *Baicalobdella*. In addition to *Cottocomephorus grewinkii*, eight sculpin hosts for *Baicalobdella* are newly recorded: *Paracottus knerii* (Cottocomephorinae), and *Abyssocottus korotneffi*, *Asprocottus platycephalus*, *Batrachocottus baicalensis*, *Batrachocottus multiradiatus*, *Cyphocottus megalops*, *Limnocottus griseus* and *Procottus major* (Abyssocottinae). These host fishes are mass infected by *Baicalobdella* leeches (up to 75%) in different sites of Lake Baikal. Comparative morphological analysis of *Baicalobdella* infecting sculpin fishes vs. *Baicalobdella torquata* proper revealed nine distinctive features that allow confirming a separate systematic position and revalidating original name *Baicalobdella cottidarum* Dogiel, 1957.

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Lake Baikal is known as one of the most ancient (25-30 million years), the deepest (1637 m), the largest (volume of 23 000 cubic km³, length of 636 km, maximal width up to 80 km, coastal line of 1800 km) freshwater reservoirs of the planet (20% affordable drinking water), with the status of Natural World Heritage Site from 1996. A unique set of the lake natural conditions has resulted in the fact that the lake has a unique complex of living organisms. Parasite fauna of Lake Baikal consists of 300 species including 28 leech species [1-4]. These species are adapted to living in cold, clean, oxygenated water and parasitize the Baikal endemic animals, mainly bullheads and amphipods. Until now, nothing was known on character and quantitative parameters of the disease of Baikal aquatic organisms caused by leeches. However, epizootics involving natural salmon fish were recorded in the adjacent territories of Eastern Siberia [5]. Leeches have a negative effect on a host health. Undoubtedly, ulceration, hemorrhage, and inflammation associated with leech attachment sites weaken the host and may predispose hosts to bacterial infections. There are evidences that leeches serve as mechanical vectors of viruses [6,7]. Leeches can transmit hematozoa including trematodes, cestodes and nematodes [8], and parasitic flagellates [9], which are considered to be pathogenic organisms for aquatic animals.

The aim of this paper is the investigation of Baikal cottoid fish bdellosis caused by *Baicalobdella* leech species and the accurate definition of the taxonomic status of the parasite. To characterize the infection, conventional parasitological indicators, such as prevalence

E-mail addresses: irina@lin.irk.ru (I.A. Kaygorodova), natalja@lin.irk.ru (N.V. Sorokovikova).

(P), intensity (I) and abundance (A) of parasites were evaluated using the Quantitative Parasitology 3.0 Program [10].

Data on bdellosis of sculpin fish in this lake were obtained during ichthyologic surveys of 2011 and 2012. Biological materials were collected by scuba divers at a depth of 2–42 m and by fishing gear at 40–1260 m.

Fish infected with leeches were found in west part of South Baikal (SB-W), east part of South Baikal (SB-E), west part of Middle Baikal (MB-W), east part of Middle Baikal (MB-E), and east part of North Baikal (NB-E), of them three localities in SB-W – Listvyanichny Bay, depth of 3-3.5 m, 51°52′05″N, 104°49′50″E, July 2011 and 2012; Listvyanichny Bay, depth of 180 m, 51°51′54″N, 104°49′27″E, March 2012; and opposite the village Bolshie Koty, depth of 2–15 m, 51°53′98″N. 105°03′83″E, July 2011 and August 2012; one locality in SB-E – opposite the village Vydrino, depth of 7 m, 51°28′85″N, 104°25′34″E, March 2011; three localities in MB-W – Maloe More Strait, depth of 3–6 m, 53°04′25″N, 106°55′48″E, May 2011; Maloe More Strait, depth of 250-300 m, 53°27′ 82″N, 107°36′51″E, June 2011; and Maloe More Strait, depth of 275-285 m, 53°22'33"N, 107°42'15"E, August 2012; one locality in MB-E – Barguzin Bay, depth of 15-32 m, 53°30′84″N, 108°32′39″E, August 2012; two localities in NB-E – Yakshakan Bay, depth of 11 m, 54°47′50″N, 109°37′16″E, August 2012, and 146 m, 54°44′05″N, 109°34′95″E, August 2012. Altogether, 1085 and 328 sculpin fish individuals were examined for leech infection in 2011 and 2012, respectively.

During the 2-year parasitological studies at Lake Baikal, we inspected 1420 demersal fish of 19 species from 10 genera for the presence of bdellosis: *Abyssocottus korotneffi* - 21, *Asprocottus intermedius* - 44, *Asprocottus korjakovi* - 51, *Asprocottus minor* - 48,







^{*} Corresponding author at: 3 Ulan-Batorskaya str., Irkutsk 664033, Russia. Tel.: +7 3952 422923; fax: +7 3952 425405.

Asprocottus platycephalus — 56, Asprocottus pulcher — 7, Batrachocottus baicalensis — 37, Batrachocottus multiradiatus — 45, Batrachocottus nikolskii — 138, Batrachocottus talievi — 39, Cyphocottus megalops — 17, Limnocottus griseus — 19, Neocottus thermalis — 9, Procottus major — 32, Comephorus baicalensis — 203, Comephorus dybowskii — 188, Cottocomephorus alexandrae — 7, Cottocomephorus grewinkii — 225, and Paracottus knerii — 234.

All studied fish belonged to the order Scorpaeniformes. Since there is no shared opinion on the division of Scorpaeniformes into families: the number ranges from 26 to 35 families [11,12], so we followed the classification system of Bogutskaya & Naseka [13], according to which the family Cottidae Bonapatre, 1831 consists of four subfamilies (Cottinae, Abyssocottinae, Comephorinae, and Cottocomephorinae) including 35 species of 12 genera inhabiting Lake Baikal. Baikal sculpins examined in this study belonged to three of these subfamilies — Abyssocottinae Berg, 1907, Comephorinae Gunther, 1861, and Cottocomephorinae Taliev, 1955. Among infected fish, there were representatives of 9 species, with 45.6% of leech infestation. Leeches were detected on 313 individuals. However, leech parasites have never been detected in the catches with abundant comephorines (altogether 391 individuals). This fact can be attributed to their unique mode of existence.

The subfamily Comephorinae includes a single genus of peculiar, sculpin-like fishes endemic to Lake Baikal [13]. There are currently two recognized species in this genus: C. baicalensis (Pallas, 1776) (big Baikal oilfish or big golomyanka) and C. dybowskii Korotneff, 1904 (little Baikal oilfish or little golomyanka). Golomyanka is a transparent fish without scales and swim bladder, with a high content of fat in muscle mass (up to 40%). These fishes are unusual because of vertical movement throughout the entire water column to maximum depths of Lake Baikal during the day. They are considered the world's most abyssal freshwater fish. Special features of these species are viviparity and cannibalism (eating their own juveniles). Golomyanka feeds mainly on zooplankton. Despite the fact that the oilfish has a solitary lifestyle, without bunching in shoals, the biomass of the fish in the lake is amounted to 200 000 tons making it one of the most populous vertebrate forms in Lake Baikal (three-quarters of the total biomass). Thus, the specific lifestyle mainly independence (or minimal dependence) of the Baikal comephorines from a bottom eliminates a possibility of infection by annelid parasites.

The Cottocomephorinae, the bighead sculpin or Baikal sculpin, is a small subfamily of scorpaeniform fishes endemic to Russia where they are found mostly in Lake Baikal and surrounding lakes and rivers, in the Yenisei River basin, and lakes in the Republic of Tuva [14]. Bighead sculpin belong to the group of freshwater benthopelagic fishes. Two of the four known Baikal species – *C. grewinkii* and *P. knerii* are prone to suffer bdellosis, with prevalence of 52% in 2011 and 55% in 2012 (Table 1). In 2012, *P. knerii* was reported to have maximal infestation with prevalence of 64%, infection intensity up to 13 leeches per fish with the 5–6 modal intervals, and abundance index of 2.88.

Nevertheless, maximal leech intensity was registered on *P. knerii* in 2011. The morbidity level of *C. grewinkii* was slightly lower compared to *P. knerii* and was 47% and 44% in 2011 and 2012 respectively. The infection intensity (I) was also lower in *C. grewinkii*, although the modal interval (MI) in both species was almost the same. Parasites were situated on fins, mainly at the base of the dorsal (Fig. 1A) and thoracic fins (Fig. 1C). Atypical leech placing was found in a spawning goby (Fig. 1B). Moreover, numerous juvenile leeches were observed feeding on the *P. knerii* eggs (Fig. 1D). A nutritional value of sculpin eggs was tested and confirmed in the laboratory.

The Abyssocottinae is a subfamily of deep-water sculpin that are entirely restricted to Lake Baikal, most of them living at depths below 200 m. There are 28 species classified into 8 genera [13]. 563 studied fishes belong to 14 species; leeches were collected from 7 of them — *A. korotneffi, Abyssocottus platycephalus, B. baicalensis, B. multiradiatus, C. megalops, L. griseus* and *P. major.* Leech prevalence in this species group was 21% in 2011 and 46% in 2012 (Table 1). High infestation was detected in *L. griseus* and *P. major* in 2012 (Table 1). *L. griseus* revealed the 100% prevalence of bdellosis in 2012. However, it was likely due to the small sample size.

The morbidity level of bdellosis was higher in cottocomephorines compared with abyssocottines (av. 53% vs. 30%, on average for 2 years). The exception is deep-water *P. major* with the highest level of infestation that was detected in 2012 (Table 1). Based on the data, we assume that the high prevalence of bdellosis in Baikal sculpin is environmentally dependent. Littoral species suffer from bdellosis more often and are more intensely affected than abyssal species. This is due to that a host-species population density at smaller depths, as a rule, is higher than in the deep parts. This hypothesis is confirmed by high prevalence of bdellosis not only in Cottocomepharinae, which are adapted to littoral zone, but also in *P. major*, one of the most abundant species in the abyssal zone of Lake Baikal.

The extensive collection of leeches obtained in this study allowed us to analyze a large sample (1193 specimens) inhabiting the different parts of the huge lake and parasitizing different sculpin species. All collected leeches have a set of characters, which is conformable to the original description of the genus *Baicalobdella* [15] since they possess the following morphological (or diagnostic) characteristics: a minor body size, a clear body division into trachelosome and urosome, presence of 11 small lateral bubbles, and 5 pairs of testisacs.

Initially, the two species belonging to this genus have been described – *Baicalobdella torquata* (Grube, 1871) and *Baicalobdella cottidarum* Dogiel, 1957 feeding on amphipods and cottoid fish respectively. The species *B. cottidarum* became to be an ambiguous taxon, since its autonomy was abolished and only *B. torquata* became a valid species parasitizing both amphipods and sculpins [16–18]. Recently, the validity of this synonymization has been questioned, but at that moment there was not a total confidence due to the deficient number of available specimens [3,4]. This is the first time, when a study is carried out on such a high amount of *Baicalobdella* samples and specimens. Therefore,

Table 1
Infection of Baikal cottoid fishes with leech <i>Baicalobdella cottidarum</i> in 2011 and 2012.

Year	Index	C. grewinkii	P. knerii	A. korotneffi	A. platycephalus	B. baicalensis	B. multiradiatus	C. megalops	L. griseus	P. major
2011	Ν	125	120	18	48	-	45	12	17	-
	Р	47%	57%	28%	23%		18%	17%	18%	
	Ι	1-8	1-20	2-4	3–8		1-2	2-4	1	
	MI	4-5	3–5	3	3		1-2	2-4	1	
	Α	1.86	2.43	0.83	0.81		0.29	0.50	0.18	
2012	Ν	100	114	3	8	37	-	5	2	32
	Р	44%	64%	0	0	38%		0	100%	75%
	Ι	1-10	1-13			1–4			2-3	1-6
	MI	3–5	5-6			1-4			2-3	3-4
	А	1.66	2.88			0.89			2.50	1.91

N - number studied fishes; P - prevalence in %; I - infection intensity; MI - modal interval of I; A - abundance index.

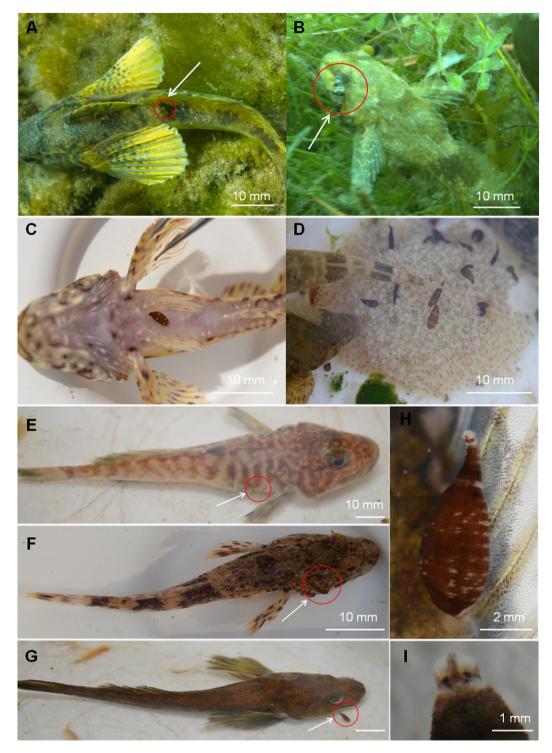


Fig. 1. Photographs of the Baikal sculpin fishes and its leech parasites. Position of leech parasites in fish is encircled and indicated by arrow. (A) *Cottocomephorus grewinkii* with leech on the base of dorsal fin. (B) Leech attached to the eye of *Paracottus knerii* (C) Leech parasite at the base of thoracic fin of *Paracottus knerii*. (D) Numerous leeches of *Baicalobdella cottidarum* on a batch of *Paracottus knerii* eggs. (E) Leech attached to *Procottus major*. (F) Leech in *Batrachocottus baicalensis*. (G) *Limnocottus griseus* with leech. Scale bar is 10 mm. (H) Intravital morphology of sculpin dependent *Baicalobdella* specimen. (I) Anterior end of leech's body with two pairs of eyes.

existing species descriptions were supplemented with intravital morphological details. The comparative analysis of leeches parasitizing sculpin fishes (this study) and their congener parasitizing amphipods (previous study) [3,4] revealed that the parasites sufficiently well diverge on external morphology as follows:

1. Although both forms are of minor sizes, sculpin's leeches are a bit larger. Their body length is up to 11 mm with a modal value of 7–8

mm, while the amphipod dependant leech is never longer than 8 mm (6–7 mm in most cases). The difference in body length (with the same body width) is 15–37%.

2. In spite of the maximal body width of both forms is identical, the difference lies in widest part location. The widest part of sculpin's leech is in the middle of urosome, while amphipod leech maximum body width is related to the posterior third of the body.

- 3. Due to different body proportions, body shape is clearly distinguishable. Unlike the spindle-shaped amphipod leech, the sculpin's leech has an ellipsoidal body.
- 4. Furthermore, sculpin's leech body has dorsiventral flattening.
- 5. Trachelosome of sculpin's leeches is almost twice shorter (0.8–0.9 mm vs. 1.5 mm) and 1.5 times wider (0.7–0.8 mm vs. 0.5 mm).
- Additionally, there are four transverse stripes of white spots on trachelosome of amphipod dependant leeches. There are no such features at sculpin's parasites.
- Another distinctive quality is the sizes of suckers. Having the same width, both suckers are twice longer in amphipod leech with -0.5 mm vs. 0.25 mm and 0.9–1.0 mm vs. 0.5 mm, anterior and posterior suckers respectively.
- 8. The most definite (pronounced) morphological character, which could be used as a key to determine these two *Baicalobdella*, is a shape of clitellum. Amphipod parasitizing leech has a ring-like milky white clitellum, whereas sculpin's leech has a dorsally interrupted half-round clitellum of the same color.
- 9. Finally, there are evident distinctions in their coloration. The urosome color of amphipod leech varies from evenly green to brown with marble white spots in contrast to sculpin dependant leech, which is usually dark brown, quite often with rhomboid white stripes.

Based on the above, it might be reasonable to speculate that in point of fact the Baicalobdella is not the monotypic genus contrary to preconceived idea [16-18], which still has its followers. A large set of fresh data enabled us to reveal complex of morphological qualities to be sufficient for distinction of two species within Baicalobdella parasitizing either sculpin (vertebrates) or amphipods (invertebrates). The meticulous parasitological examination of Baikal sculpin showed that infection cases with leeches of *B. torquata* proper morphotype had never been detected among numerous tested fish. A plausible explanation for species division is via distinct ecological niches and adaptation to different hosts. A consequence of this adaptation is forming of the unique complex of distinctive features of external morphology in each ecogroup and as a result a split or divergence of species. Hence, the combination of morphological differences and distinct hosts makes possible a rehabilitation of parasite species name *B. cottidarum* Dogiel, 1957 for leeches parasitizing Baikal sculpin fish.

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