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New information on the distribution pattern of *Acanthobdella peledina* (Annelida, Acanthobdellida) in Eastern Siberia

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The Acanthobdellida is a group of annelid parasites of fish, which are restricted to subarctic parts of the Northern Hemisphere. These ancient worms arouse the interest of the scientific world due to their mosaic combination of (1) primitive oligochaetous characters (e.g. the presence of setae on the very anterior segments, a lateral nerve system and a metameric coelomic cavity around the intestinal canal) and (2) evolutionarily advanced hirudinean ecological and morphological features (e.g. suckers and parasitic lifestyle), attesting to their intermediate role between Oligochaeta and Euhirudinea (i.e. leeches with setae).

The first time these leech-like fish parasites were found was on *Coregonus peled* (Gmelin, 1789) of the Lower Yenisei River during Middendorff's expedition and they were described in 1851 by the German zoologist A. Grube (Grube 1851). Two species are currently known within the group. *Acanthobdella peledina* Grube, 1851 is one of them. Our basic knowledge of the morphology of *A. peledina* goes back to Livanow's monograph of 1905, where this species was identified as an ancient hirudinean; close phylogenetic relationships to the leeches and to the oligochaetes were suggested (Livanow 1905). A series of later investigations corroborated Livanow's phylogenetic hypothesis on the transitional position of the "living fossil" *A. peledina* between oligochaetes and leeches, as shown by morphological and life cycle investigations (Franzén 1991, Purschke *et al.* 1993, Brinkhurst 1999, Borda & Siddall 2004) as well as by molecular phylogenetic analyses (Trontelj *et al.* 1999, Siddall *et al.* 2001, Kaygorodova & Sherbakov 2006).

These archaic freshwater leech-like annelids of the genus *Acanthobdella* are semi-permanent parasites feeding exclusively on salmonids. In contrast to its closest relative *Paracanthobdella livanowi* (Epstein, 1966), whose habitation is limited to fresh waters of the Kamchatka and Chukotka peninsulas (Epstein 1966), *A. peledina* inhabits high latitudes of the Northern Palearctic, from Norway on the west to the Kolyma Region on the east (Lukin 1976), and it is likely found in North America according to a few evidences (Holmquist 1974, Hauck *et al*. 1979). Environmentally, *A. peledina* is a psychrophilic (cold-loving) stenobiont (living under narrow and relatively constant ambient conditions) of exclusively oligotrophic waters of the northern mountain rivers and lakes. Its distribution is much more restricted compared with that of its hosts – Salmonidae, Coregonidae and Thymallidae (Pronin 1979, Matveev & Pronin 2010), i.e. *A. peledina* is more stenoecic than the fishes it infests. Since acanthobdellids have usually a relatively low frequency of infestation (4–5 %), it is obvious that the total abundance of certain populations may be very low in the Siberian aquatic ecosystems. Therefore, despite a wide range of potential habitats, the acanthobdellids are classified as rare species and listed as endangered in the Republic of Buryatia and Irkutsk Region.

The relatively recent discoveries of *A. peledina* on salmon fishes of the Kuando-Chara watershed and on an arctic char in Lake Frolikha (Pronin 1971, Pronin 1979), graylings of the Upper Angara River (Baikal basin) (Pronin 1971), and on other salmonids from the Chechuy, Rassokha and Chaya rivers (Lena basin) (Matveev & Pronin 2010, Kaygorodova *et al.* 2012), defined the southern boundary of its distribution: the hydro-geographic system of the Baikal and Stanovoy Uplands with Lake Frolikha as the most southern location, i.e. N55.5° (Pronin 1979). This remote area separated by several thousands of kilometers from its present and known distribution in the subarctic zone of Eurasia is considered as a potential refugium of *A. peledina* (Pronin 1971, Matveev & Pronin 2010). However, during the ichthyologic survey in the western part of the Angara River basin (Fig. 1A), a massive *Acanthobdella* infestation of salmonid fish was detected randomly and this for the first time in the area. We analysed the samples collected by Dr. Elena Dzyuba on 22 and 23 October 2015 from the Iya River (53°49′0091″N, 99°35′1841″E) and in its tributary the Barbitay River (4 km upstream of the river mouth: 53°48′1351″N, 99°33′4651″E). Fishes during their feeding period

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were caught using gill nets (30–32 mm mesh) with a total length of 30 m.

We examined 325 graylings, i.e. 203 specimens from the Iya River and 122 specimens from the Barbitay River, for the presence of acanthobdellosis. Among the infected fish, we found only representatives of *Thymallus baicalensis* (Dybowski, 1874), popularly known as Baikal black grayling. Leeches were detected on 63 individuals, with total grayling lesion of 19.4%. The prevalence of acanthobdellosis was 28.08% in the Iya River whereas in the Barbitay River it was within the normal range without exceeding 5%. The infection never exceeded one leech per individual fish. Acanthobdellids were found on both male and female fish with a length of 22–33 cm, and with a sex ratio (M/F) of 3/3 and of 31/26 in the Barbitay and the Iya River, respectively.

The maximum age and length of *Thymallus baicalensis* was 10+ years and 340 mm, respectively, and its maturation was at 3–5 years (Matveev & Samusenok 2009). Parasitological studies showed that Baikal black graylings from the Angara River basin were 16.0–33.0 cm long. Only younger age groups of mature fishes suffered from acanthobdellosis: 4 three-year-old and 2 four-year-old individuals from the Barbitay River; 6 three-year-old, 36 four-year-old and 15 five-year-old individuals from the Iya River. According to the modal number, predominantly four-year-old graylings of both sexes suffered from this kind of bdellosis.

The ectoparasites were identified morphologically as belonging to *Acanthobdella peledina*. All the 63 specimens had the species-specific pattern of morphological features (cf. Livanow 1905, Dahm 1962, Kaygorodova *et al.* 2012). The length of the worms ranged from 16 to 23 mm and 4–7 mm in width. The body shape varied only slightly. Living specimens had a vermiform shape and a cossack-green (khaki) coloration with a dark transverse stripe in the middle of each somite (Fig. 1B). The three pairs of underdeveloped dark-red eyes were hardly distinguishable in living individuals and almost invisible in fixed specimens because of the rapid bleaching of the pigment. The parasites had an elongated anterior, tapering more strongly to the anterior sucker, which was not well developed. The five segments of the head portion were not separated from the adjacent body part as opposed to its single known relative *P. livanowi*, which has an obviously separated head and a rather developed anterior calycine sucker (Epstein 1966). The examined individuals had 40 setae on the first ten annuli (i.e. the first five segments); that is one of the main taxonomically significant archaic attributes of these ancient parasites, linking them to the Oligochaeta. These setae are grouped into 4 bundles of 2 setae in each segment. A similar seta apparatus is present in the oligochaete family Lumbriculidae. In contrast to lumbriculids, all setae of *A. peledina* are dark brown, hook-shaped and their location is restricted to the anterior sucker. The posterior end of the body is extended or is retracted inward (Fig. 1B). The combination of these attributes leaves no doubt of the taxonomic status of this parasite.

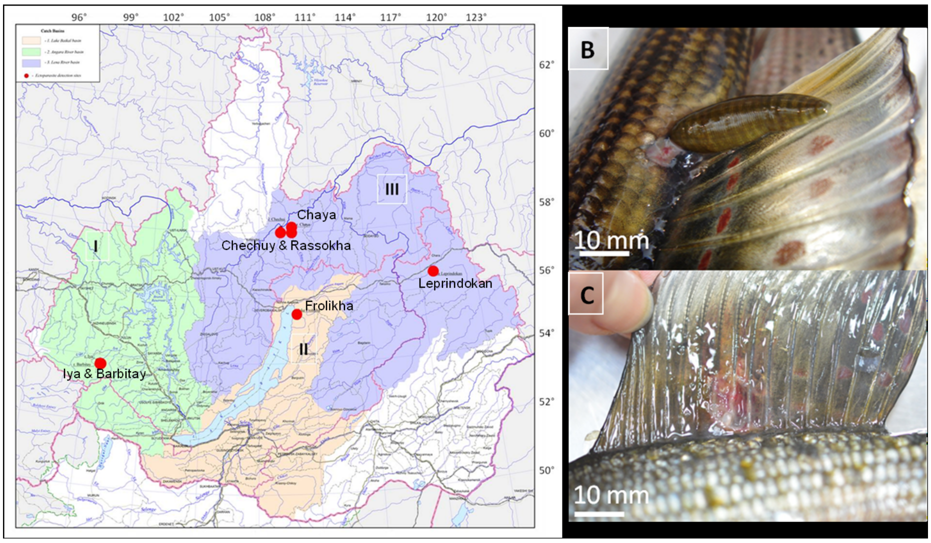

FIGURE 1. *Acanthobdella peledina* of East Siberia. (A) The map of East Siberia with the exact geographic locations of the occurrence of *A. peledina* (highlighted with red dots), including the Angara River basin (green), the Lake Baikal basin (pink) and the Lena River basin (violet). (B) External appearance of the leech-like ectoparasite *A. peledina* biting on the dorsal fin of a Baikal black grayling. (C) The wound left after a bite: a lot of tissue disappeared from the dorsal fin.

Moreover, *A. peledina* was registered for the first time in the catchment basin of the Angara River, increasing our knowledge of its distribution. To date, the Barbitay River is the most southerly area of detection of this acanthobdellidan parasite. The question arises about the so called refugium/relict aspects of the southern populations of *A. peledina* (Pronin 1971, Matveev & Pronin 2010): it is quite possible that the separation of the southern populations from the subarctic is an artefact, due to the rather difficult accessibility of the regions between and, consequently, their insufficient exploration. However, the fragmentation of its southern area is likely related to anthropogenic impacts. The insular mode of the species distribution in Eastern Siberia should be considered as debris of a former continuous area of this species in the Pleistocene.

During the study we noted that the parasites were situated on the fins, mainly at the base of the dorsal fin, forming a cluster of deep ulcerous wounds in the soft tissues of their host (Fig. 1B and C). We reaffirmed that the ectoparasites are attached to the fish skin with the setae located near the anterior sucker, and then feed on blood and tissue of their vertebrate host (Livanow 1905). Our observations support the hypothesis of epidermis feeding rather than bloodsucking. *Acanthobdella* individuals leave round wounds on the grayling skin (Fig. 1C), which are supposed to cause bacterial or fungal infections, increase stress which may lead to fish mortality.

From the second half of the 20th century onwards, there is a tendency towards a decrease in abundance and, as a consequence, a significant reduction of the distribution area of this so unique animal species *A. peledina*, including its complete extinction in some rivers and lakes of Sweden, Karelia and the Kola Peninsula because of the disappearance of its obligate hosts and the eutrophication of water bodies (Dahm 1962, Pronin 1997). The importance of these "living fossils" as a transitional form in the evolution of annelids can hardly be overestimated. Therefore, all new information on the biology and distribution of *A. peledina* is of great value in understanding its role in the evolution of the Annelida.

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