# New *Stylodrilus* species (Annelida; Clitellata; Lumbriculidae) from Lake Baikal, East Siberia

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A new oligochaete worm, *Stylodrilus aclotudi*, has been reported from the southern part of Lake Baikal in East Siberia, Russia. The new species differs in extremely long tubular atria extending as far as segment XV, that is a unique character of the genus. This new finding increases the number of *Stylodrilus* species which are endemic to the lake up to 11, confirming the existence of Baikal species flock.

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## Introduction

The freshwater oligochaete genus *Stylodrilus* Claparède, 1862 is a large and heterogeneous genus that harbors 33 species to date. Although the genus has a Holarctic distribution, except *Stylodrilus heringianus* Claparède, 1862 (a peregrine species recorded in the Australasian region and assumed to have been introduced by shipping; Brinkhurst & Jamieson, 1971), the majority of species, namely 28, are exclusive to the Palaearctic region (Martin et al., 2007). Within the confines of the single restricted unique ecosystem of Lake Baikal, the genus underwent an intense evolutionary radiation, giving rise to a species flock: among 12 species known so far, 10 are endemic to this lake.

The revision of the genus Stylodrilus (Brinkhurst, 1965) led to including *Bythonomus* Grube, 1879. The synonymy of these genera did not receive undivided support among specialists. Hrabc (1970) maintained that Bythonomus is separable from Stylodrilus by virtue of the difference in setae (simple-pointed vs. bifid), posterior blind-ending lateral blood vessels and the position of the opening of the vasa deferentia into the atria. Cook (1975) proposed to recognize a useful subgeneric category for Bythonomus that includes those species with simple-pointed setae and absence of copulatory structures. Brinkhurst & Wetzel (1984) demonstrated that the blood system and the opening of vasa deferentia are unreliable characters and do not justify keeping this separation. Rodriguez & Coates (1996) considered that setal morphology is no longer a definitive character for generic rank in the family.

Since 1965, the genus *Stylodrilus* has been enriched twice, only for Lake Baikal seven new species have been discovered and described (Hrabc, 1982; Semernoy 2004), and two species *S. parvus* and *S. mirus* have been for the first time recorded in the northern part of the lake (Snimschikova, 1987).

The present finding of a new *Stylodrilus* species gives an interesting perspective to revert to the question of the validity of the subgenera *Stylodrilus* and *Bythonomus* since all *Stylodrilus* species inhabiting Baikal except *S. mirus* (not endemic sp.) possess bifid setae, a feature shared by the present new *Stylodrilus* species, suggesting that this setal character could be more conservative than usually assumed.

## Material and methods

The material was sampled by one of the authors (IK) in the southern basin of Lake Baikal during the expedition in late September 1997. Samples were taken with a net (250 µm mesh size) handled *in situ* by a scuba diver. All samples were preserved in 80% ethanol for further molecular study. Specimens used for classical morphological work were later fixed with 7% neutralized formalin before being dissected. Worms were stained with alcoholic carmine, mounted in Canada balsam and examined under a Reichert (Austria) microscope.

Drawings were made by means of a camera Lucida. The type series are deposited in Limnological Institute, Siberian Branch of the Russian Academy of Sciences (LIN SB RAS, Irkutsk, Russia), under numbers B97.80.21 and B97.80.25.

Segments were designated by Roman numerals and septa were indicated by Arabic numbers of the adjacent segments (e.g. septum 4/5 is between segments IV and V).

Phylum Annelida

Classis Clitellata Michaelsen, 1919

Subclassis Oligochaeta Grube, 1850

Lumbriculidae Vejdovsky, 1884

Genus Stylodrilus Claparède, 1862

# Stylodrilus aclotudi sp. n.

(Fig. 1)

*Holotype.* **Russia**, Lake Baikal, southern basin, Murino Bank, depth 18 m; sandy sediment with big separate stones obstructing dredging or grabbing, 25-27.IX.1997, slide no. B97.80.21 (I. Kaygorodova), deposited in the Limnological Institute SB RAS, Irkutsk. Sexually precopulative specimen, incomplete (first 18 segments), anterior ends (head

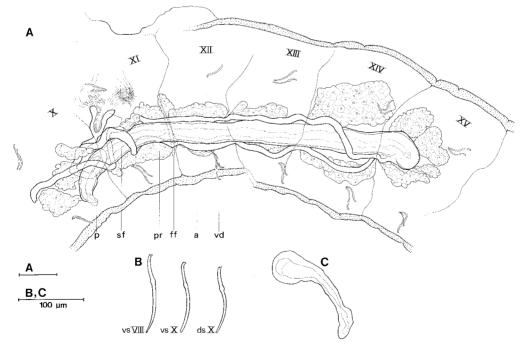
and sexual segments) cut along the midsagittal plane, tail left as is, stained in alcoholic carmine and mounted in Canada balsam.

*Paratypes*. One sexually precopulative mature specimen from the same site, incomplete (first 19 segments) dissected and mounted in the same way as the holotype; LISB slide B97.80.25.

*Etymology.* An "aclot" is, in the Walloon language, an inhabitant of Nivelles (Belgium), the town where the second author was born. "Aclotudi" (pronounced as in Latin) literally means "aclot always" in Walloon and is seen here as a homage of the second author to his native town.

Description. Medium-sized species (incomplete specimen, first 18 segments, 3.26 mm; paratype 19 segments,  $3.1 \text{ mm} \log p$ ,  $430-470 \mu m$  at widest (clitellum). Prostomium rounded. Secondary annulation from V onwards, narrow annulus anterior. Clitellum inconspicuous, approximately extending between IX and narrow annulus of XII. Two setae per bundle, slightly bifid with upper tooth reduced, sigmoid with ectal nodulus on the upper third of seta, similar in dorsal and ventral bundles, 48-78 µm long, 2.5-3.4 µm thick (Fig. 1B). Setal length increasing from 46 µm in II to 78 µm in VIII, slightly decreasing to a nearly constant length (65-71 µm) in postclitellar segments.

Dorsal pad of pharynx extending from II to III, pharyngeal glands not conspicuous on dissected



**Fig. 1**. A-C – *Stylodrilus aclotudi* sp. n. A – Right lateral view of genitalia (incomplete) in segments X-XV (holotype). B – Somatic setae (holotype). C – Left lateral view of spermathecae from segment IX (paratype LISB 97.80.25. Abbreviations: a – atrium, ds – dorsal seta, ff – female funnel, p – penis, pr – prostate gland, sf – sperm funnel, vd – vas deferens, vs – ventral seta.

Character	S. sub- carpaticus	S. longi- atriatus	S. califor- nianus	S. miran- dus	S. mol- lis	S. elon- gatus	S. aclotudi <b>sp. n.</b>
Blind posterial lateral blood vessels	Bifurcated apically	With many blind appendices	Absent	Absent	Absent	Absent	Absent
Setae	Simple- pointed	Simple- pointed		Bifid	Bifid	Bifid	Bifid
Size of setae, µm	?	154-172	48-70	73-105	42-98	200-250	48-78
Second annulation	From V	From VI	Absent	From IX	From V or VI	From VII	from V
Form of atria	Tubular	Ovoid	Tubular	Tubular	Tubular	Tubular	Tubular
Size of atria	X-XIII	X-XI	Х	Х	Х	Х	X-XV
Location of vasa deferentia connection to atrium	Proximal	Subapical	Apical	Proximal	Subapical	Subapical	Subapical
Penes	Absent	Conical	Bulbous	Thick muscular eversible	Bulbous	Small	Small

Table. Some systematic characters of *Stylodrilus* species closely related to *S. aclotudi* sp. n.

material. Two pairs of sperm sacs associated with each testis segment; anterior sperm sacs as forwards bulges of septa 8/9 or confined in X, respectively, posterior sperm sacs as backwards bulges of septa 10/11 and extending to XV. Egg sacs absent in types.

One pair of spermathecal pores opening on setal line, just behind ventral setae. Spermathecae not fully developed, restricted to IX (Fig. 1C). Spermathecal ducts tubular, slightly bulbous ectally (near spermathecal pores), as long as ampullae; ampullae small, ovoid, sperm absent.

Male pores paired, opening ventrally on setal line, behind ventral setae, in the middle of large annulus of X. Two pairs of testes in IX and X, attached ventrally to 8/9 and 9/10, respectively. Atria tubular, 1018 µm long, 61.4 µm wide, with narrow lumen, extending backwards to XIV-XV; muscle layer 5.8 µm thick (Fig. 1A, a). Proximal part of atria opening externally by means of small, droplike external penes (Fig. 1A, p). Prostate glands discrete, covering the entire atrium in bundles, separately opening onto atrium by means of short ducts (Fig. 1A, pr). Two pairs of sperm funnels, on septa 8/9 and 9/10 (Fig. 1A, sf), both directed forwards (posterior sperm funnel illustrated in Fig. 1A artefactually directed backwards due to dissection). Vasa deferentia 17-24 µm wide, more or less uniform all through their length, narrower near junction with atrial ampulla ( $12 \mu m$ ), making some loops in X before joining atrium proximal to this, running along atrial ampulla besides its muscle coat and penetrating, side by side, atrium subapically (Fig. 1A, vd).

Female pores paired, in line with setae and spermathecal and male pores. Female funnels ventrally attached to septum 11/12, opening onto intersegmental furrow XI/XII (Fig. 1A, *ff*). Ovaries paired, fixed ventrally near septum 10/11, weakly developed in types, restricted to XI.

*Remarks*. This new species belongs to the large and heterogeneous genus Stylodrilus Claparède, 1862 defined by a semi-prosoporous arrangement of male ducts, male pores in X, spermathecae in IX, and atrial musculature generally consisting of a single, more or less circular layer (Fend, 2005). Thirty-three species are presently recognized as valid, 15 have simple setae, 2 have unusual setae (corolla of thin teeth or serrate pecten of thin denticles instead of an upper tooth) and 16 have bifid setae with small upper tooth. Among the latter group, S. aclotudi sp. n. is easily distinguished by having non-petiolate atria uniquely elongated, extending as far as XV (Table). S. californianus Rodriguez, 1996, S. elongatus Semernoy, 2004 (non Semernoy, 2001), S. mirandus (Hrab•, 1982) and S. mollis Timm, 1998 have more or less tubular or cylindrical atria but they are restricted to

the male segment. S. mirandus is distinctly different by its thick muscular eversible penis (Hrabc, 1982; Semernoy, 1982). In contrast to S. aclotudi sp. n., both S. californianus and S. mollis have atria opening through bulbous penes, extending through the male pore either as a papilla or as an inconspicuous soft stick. The latter species is in need of re-description since illustrations of genital organs in paratypes clearly show differences that should put these specimens in different species (notably the atrial form and relative position of spermathecal pores to dissepiment). The Baikal S. elongatus is morphologically the closest to S. aclotudi **sp. n.** Like the latter, it has bifid setae. non-petiolate tubular atria, vasa deferentia that join atria subapically and small penes; atria are however restricted to the male segment, secondary annulation starts from VII onwards and size of setae is noticeably larger (in three times as minimum).

Only two *Stylodrilus* species such as *S. sub-carpaticus* (Hrabe, 1929) and *S. longiatriatus* Dembitsky, 1976 possess long atria which are outside the segment limits with male pores (Table). Moreover, *S. subcarpaticus* also has tubular atria. However, unlike *S. aclotudi* **sp. n.**, these both species have much shorter atria (X-XIII and X-XI correspondingly) and belong to the group with simple-pointed setae.

*Geographical distribution and habitat.* To date, only known from type locality.

## Discussion

Today, 14 Stylodrilus species are known from Lake Baikal: S. aclotudi sp. n., S. asiaticus (Michaelsen, 1901), S. contractus Semernoy, 2004, S. crassus (Isossimov, 1948), S. elongatus Semernoy, 2004, S. gracilis Semernoy, 2004, S. insperatus Semernoy, 2004, S. minutus Hrabc, 1970, S. mirandus (Hrabc, 1982), S. mirus (Cekanovskaya, 1956); S. opisthoannulatus (Isossimov, 1948), S. parvus (Hrabc and Cernosvitov, 1927), S. subitus Semernoy, 2004 and S. sulcatus Semernoy, 2004.

All of them are endemic to the lake except three: *S. mirus*, *S. opisthoannulatus* and *S. parvus*. Though *S. opisthoannulatus* is commonly represented in Lake Baikal at different depths (2-860 m), it was also found in the Angara reservoirs, the Yenisei River and Yenisei Gulf (Chekanovskaya, 1956), where this species could most likely have come from Baikal through the Angara River, the only effluent of the lake flowing into the Yenisei.

*S. parvus* was also mentioned in the Balkans and ground water in France It is hence tantalising to consider *S. parvus* as the closest to the ancestor species that colonized the lake and gave rise to the *Stylodrilus* species flock, suggesting that the absence of penes, spherical atria, bifid setae and vasa deferentia that join atria subapically are plesiomorphic characters within the flock. However, it is worth noting that Semernoy (2001, 2004) omitted this species in his recent updated species list of Lake Baikal, implicitly suggesting, without formal substantiation, that *S. parvus* was irrelevant in Lake Baikal. It is also possible that the present-day group of Baikal *Stylodrilus* species derives from another ancestral form, or results from multiple, independent invasions of species that subsequently radiated into the lake. Molecular tools should help solve this issue.

As regards *S. mirus*, it was originally described from the Lower Yenisei (Chekanovskaya, 1956). Snimschikova (1994) indicated *S. mirus* in Lake Baikal but the first species determination (Snimschikova, 1987) was mentioned in Northern Baikal with a question mark, leaving some doubt about the validity of this record. Semernoy (2001, 2004) did not include this taxon in his species list. We accept his decision as a conservative measure. Recently Kaygorodova (2000) has found this species in the southern part of Baikal. The later molecular phylogenetic analysis (Kaygorodova et al., 2007) verified the identification of this species.

In spite of this, it is worth noting that all Baikal species belong to the Stylodrilus group that possesses bifid setae with reduced upper tooth, except S. mirus, a species with simple-pointed setae. S. insperatus and S. subitus have unusual setae with duplicated upper teeth but it may be supposed that these features are derived from an ancestral bifid state of setae, with a reduced upper tooth. Recent molecular studies (Kaygorodova, 2000; Kaygorodova & Sherbakov, 2006; Kaygorodova et al., 2007) revealed that the majority of these Stylodrilus species originated within the lake, as a result of local speciation starting about 3 million years ago. This suggests that the character "simple-pointed / bifid setae" is not as variable in this genus as usually assumed, or observed in other genera (see Rodriguez & Coates, 1998), at least within the evolutionary time scale considered (about 3 Myr). Although setal characters are usually considered as not very reliable in phylogenetic analyses of oligochaetes (Erséus, 1987), it is not unreasonable to assume a low degree of homoplasy of this character in *Stylodrilus sensu lato*. In this case, it would give more consistency to a distinction of two Stylodrilus groups based on the possession of simple-pointed or bifid setae, although both groups are probably very closely related. As a result, the proposed subgeneric rank for Bythonomus, including all those species with simple-pointed setae, (Cook, 1975) would appear to be justified.

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