

## Appearances of the non-indigenous *Helobdella europaea* Kutschera, 1987 (Hirudinea, Glossiphoniidae) in Hungarian watercourses

KRISTÓF MÁLNÁS, KRISZTIÁN KOVÁCS, MÁRK FICSÓR, PÉTER JUHÁSZ, ZOLTÁN MÜLLER, PÉTER OLAJOS & BÉLA KISS

**ABSTRACT:** Mass occurrence of non-indigenous *Helobdella* species was detected in the highly modified, channelized Tóció-stream near the city of Debrecen (E Hungary). The specimens found were determined as *Helobdella europaea* Kutschera, 1987. Similar specimens belonging also to the *Helobdella triserialis* species-complex were collected earlier near Hévíz and Keszthely (W Hungary) in 2009. Despite the mass occurrence, the *H. europaea* specimens disappeared by the winter of 2016, similarly to the former occurrence of the species complex in W Hungary.

### Introduction

The leech *Helobdella europaea* Kutschera, 1987 was first described as *Helobdella striata* Kutschera, 1985 from Germany (KUTSCHERA 1985) and was considered as the second European *Helobdella* species. Apart from some intermittent German populations (KUTSCHERA & WIRTZ 1986, PEIFFER et al. 2004), *H. europaea* was found in the Netherlands (HAAREN 2004), Spain (REYES-PRieto 2013), in New-Zealand, Australia and Hawaii (GOVEDICH & DAVIES 1998 – referred to as *H. papillornata* Govedich & Davies, 1998; SIDDALL & BUDINOFF 2005), and in Taiwan, South-Africa and California (SIDDALL & BUDINOFF 2005, BELY & WEISBLAT 2006, LAI & CHEN 2009).

*Helobdella europaea* belongs to the ambiguous *H. triserialis* species-complex which has a Neotropic distribution. Studies of mitochondrial DNA sequences suggest that the species-complex is represented by morphologically similar but taxonomically separate leeches (BELY & WEISBLAT 2006). Thus, the discrimination of *H. europaea* and the closely related *H. triserialis* (Blanchard, 1849) is quite problematic (UTEVSKY & MAZEPA 2004, LAI & CHEN 2009). Although the exact geographical range is still unknown, *H. europaea* is also presumed to originate from South-America (KUTSCHERA 2004). In any case, *H. europaea* is a non-indigenous species in Europe, which presumably spreads via aquarium trade (KUTSCHERA 2004).

The occurrence of *Helobdella europaea* in Hungary has not been reported to date. Here we present the first Hungarian records of the South-American *H. europaea* along with a description of the habitats in which it was found.

### Materials and methods

Obviously unfamiliar leeches were collected in surveys in the area of Keszthely (W Hungary) in 2009 and in the area of Debrecen (E Hungary) in 2015. The leech specimens were found both in faunistical surveys and semi-quantitative samples collected according to the Sampling Protocol of the Hungarian National Biodiversity Monitoring Programme ([www.termeszetvedelem.hu](http://www.termeszetvedelem.hu)), and the sampling protocol of the Hungarian Water Framework Directive (MSZ EN

27828: 1998). The first collected specimens were preserved in 70% ethanol. After the identification of the first specimens in 2015, further specimens were collected in 2015 and preserved in 90% ethanol for mitochondrial DNA comparison. Living specimens were also collected and subsequently kept in an aquarium for further observation.

The identification of the leeches was carried out on dyed and sliced specimens primarily based on the keys of NESEMANN & NEUBERT (1999) and additionally KUTSCHERA (1987) and LAI & CHEN (2009). The identification was checked and verified by mitochondrial DNA sequence analysis by Ulrich Kutschera.

## Results and discussion

Leeches belonging to the *Helobdella triserialis* species-complex were collected from four separate watercourses in Hungary. In 2009, three specimens were collected in the area of Hévíz: one from Hévíz stream and two from the Páhoki Channel. Despite additional targeted searches, we found no further specimens in these streams since 2009, thus, we concluded that these populations are probably intermittent.

In August of 2015, several specimens were found in the Tóció-ditch near Debrecen, and about two months later in an adjacent section of the Tóció-stream between Debrecen and Mikepércs. The specimens collected in the Debrecen area were determined to be *Helobdella europaea* Kutschera, 1987 based on external and internal characteristics (KUTSCHERA 1987, LAI & CHEN 2009). The leeches collected had morphological traits that suggested that the specimens belonged to the *Helobdella* genus (flattened body, one pair of eyes, one segment distance between the gonopores). The dorsal surface of the living leech had dark longitudinal stripes on a brownish, mottled white basis. The brownish pattern faded on specimens preserved in alcohol. Dark pigmented prominent papillae were arranged in five longitudinal rows. On the rear body, additional papillae can rise. The median row of papillae is attended by conspicuous median lines. On sliced specimens the 5 pairs of caeca were observable in the correct order (see: KUTSCHERA 1987, LAI & CHEN 2009). The identification was verified by mitochondrial DNA sequence analysis in this case. In contrast, the identification of specimens collected in 2009 was based entirely on external morphology (KUTSCHERA 1987, NESEMANN & NEUBERT 1999). Those specimens were preserved in 70% alcohol, so they were inappropriate for DNA sequence analysis. Thus the exact taxonomical classification in the *H. triserialis* species-complex is not known, and those specimen also can be *H. triserialis*.

The living leeches kept in the aquarium were fed with Tubificid worms and survived for several months. Feeding on *Physella acuta* was also observed. The leeches were observed also on the shell of living *Lymnea stagnalis* and *Planorbarius corneus* but in these cases the actual feeding was not confirmed. The *P. acuta* specimens attacked by the leeches usually died subsequently. Obviously, the dead bodies of bigger snail species (*P. corneus*, *L. stagnalis*) also were attractive for the leeches. Feeding on *Asellus aquaticus* was not observed.

### Description of the habitat

The rivulets inhabited by the newly found *Helobdella* leeches are heavily modified slow flowing lowland rivulets or artificial channels. The Páhoki Channel and the Hévíz stream are about 3-4-m wide and about 1-m deep channels covered by dense macro-vegetation. Both rivulets carry some water from the thermal baths of Hévíz. The studied section of the Tóció-stream is also a heavily modified channel that drains the purified sewage of the city of Debrecen. As a consequence of a recent removal of the vegetation, the riverbed was mostly bare,

and only small areas were covered with *Potamogeton nodosus* and *P. pectinatus*. The Tóció Ditch is also a rainwater-draining channel that crosses the area of the Debrecen Airport before joining the Tóció-stream. The whole, c. 500-m-long section is covered by emergent reed vegetation. Our findings thus confirm that *H. europaea* prefers channelized slow-flowing streams and drainage ditches (HAAREN 2004, LAI & CHEN 2009).

### Spread of the population

In the area of Debrecen, only five specimens of *Helobdella europaea* were found in the Tóció-ditch and there were none in the Tóció-stream in August, 2015. However, in October, dozens of *H. europaea* specimens were collected from a 5.5-km-long section of the Tóció-stream (between 47°29'30.48"N, 21°35'50.84"E and 47°26'36.23"N, 21°36'1.84"E). During the surveys, many leeches attached themselves to the waders or to the net. Moreover, it was the most abundant leech at the section near the airport. It appeared that *H. europaea* was newly introduced to the Tóció-ditch, and by October it spread to the Tóció Stream to become the most dominant Glossiphoniid-species. Probably as a consequence of the recent cleaning of the Tóció-stream, it could spread swiftly and without any difficulty. As it happened in the Hévíz cases, the population collapsed during the winter of 2016, and no further specimens were found ever since.

### List of data

*Helobdella europaea* Kutschera, 1987 – Debrecen: Tóció, N 47.459529°, E 21.598811°, 26.10.2015, 18, K. Málnás; N 47.465240°, E 21.597374°, 26.10.2015, 23, K. Málnás; N 47.478965°, E 21.597742°, 26.10.2015, 10, K. Málnás; N 47.491805°, E 21.597457°, 26.10.2015, 35, K. Málnás; Tóció-árok, N 47.488935°, E 21.603918°, 14.08.2015, 1, K. Málnás; N 47.49468°, E 21.61086°, 01.09.2015, 30, K. Málnás (These specimens are deposited in the Mátra Museum of Hungarian Natural History Museum, Gyöngyös; MM: A2016-1); N 47.494079°, E 21.610553°, 14.08.2015, 4, K. Málnás. – Mikepércs: Tóció, N 47.443476°, E 21.600307°, 28.10.2015, 2, K. Málnás.

*Helobdella triserialis* species-complex\* – Keszthely: Hévíz-folyás, N 46.766387°, E 17.208993°, 02.07.2009, 1, P. Olajos. – Alsópáhok: Páhoki-övecsatorna, N 46.759798°, E 17.204931°, 24.09.2009, 2, K. Kovács.

\*In the absence of DNA sequence analysis, an unambiguous identification was not possible.

**Acknowledgement:** We wish to express our sincerest gratitude to Prof. Dr. Ulrich KUTSCHERA (Institut für Biologie, Universität Kassel, Kassel) for the verification of our identification with mitochondrial DNA sequence analysis and we thank Eszter Á. KRASZNAI and Szabolcs LENGYEL (Danube Research Institute, Department of Tisza River Research, Debrecen) for their linguistic support.

### References

- BELY, A. E. & WEISBLAT, D. A. (2006): Lessons from leeches: a call for DNA barcoding in the lab. – *Evolution & Development*, 8(6): 491–501.
- GOVEDICH, F. R. & DAVIES, R. W. (1998): The first record of the genus *Helobdella* (Hirudinoidea: Glossiphoniidae) from Australia, with a description of a new species, *Helobdella papillornata*. – *Hydrobiologia*, 389: 45–49.
- HAAREN VAN T., HOR, H., SOES, M. & TEMPELMAN, D. (2004): The freshwater leeches (Hirudinea) of The Netherlands. – *Lauterbornia*, 52: 113–131.
- [http://www.termeszetvedelem.hu/\\_user/downloads/mintavetel/makrozoo\\_protokoll\\_uj\\_080218.pdf](http://www.termeszetvedelem.hu/_user/downloads/mintavetel/makrozoo_protokoll_uj_080218.pdf)
- KUTSCHERA, U. (1985): Beschreibung einer neuen Egelart, *Helobdella striata* nov. sp. (Hirudinea: Glossiphoniidae). – *Zoologische Jahrbücher Systematik*, 112: 469–476.
- KUTSCHERA, U. (1987): Notes on the taxonomy and biology of leeches of the genus *Helobdella* Blanchard 1896 (Hirudinea: Glossiphoniidae). – *Zoologischer Anzeiger*, 219: 321–323.

- KUTSCHERA, U. (2004): The freshwater leech *Helobdella europaea* (Hirudinea: Glossiphoniidae): an invasive species from South America? – *Lauterbornia*, 52: 153–162.
- KUTSCHERA, U. & WIRTZ, P. (1986): Reproductive behavior and parental care of *Helobdella striata* (Hirudinea: Glossiphoniidae): a leech that feeds its young. – *Ethology*, 72: 132–142.
- LAI, Y. T., CHANG, C. H. & CHEN, J. H. (2009): Two new species of *Helobdella* Blanchard, 1896 (Hirudinida: Rhynchocdellida: Glossiphoniidae) from Taiwan, with a checklist of Hirudinea fauna of the Island. – *Zootaxa*, 2068: 27–46.
- MSZ EN 27828 (1998): Vizminőség. Biológiai mintavétel. A vízi bentikus makroszkópikus gerinctelenek kézhálós mintavételének irányelvei (ISO 7828:1985). [Water quality – Methods of biological sampling – Guidance on handnet sampling of aquatic benthic macro-invertebrates.] pp.11.
- NESEMANN, H. & NEUBERT, E. (1999): Annelida, Clitellata: Branchiobdellida, Acanthobdellea, Hirudinea. – *Süßwasserfauna von Mitteleuropa*, Band 6/2. Spektrum Akademischer Verlag, Heidelberg, Berlin, 1–178.
- PFEIFFER, I., BREINIG, B. & KUTSCHERA, U. (2004): The occurrence of an Australian leech species (genus *Helobdella*) in German freshwater habitats as revealed by mitochondrial DNA sequences. – *Molecular Phylogenetics and Evolution*, 33: 214–219.
- REYES-PRieto, M., OCEGUERA-FIGUEROA, A., SNELL, S., NEGREDO, A., BARBA, E., FERNÁNDEZ, L., MOYA, A. & LATORRE, A. (2013): DNA barcodes reveal the presence of the introduced freshwater leech *Helobdella europaea* in Spain. – *Mitochondrial DNA*, 25(5): 1–7.
- SIDDALL, M. E. & BUDINOFF, R. B. (2005): DNA-barcoding evidence for widespread introductions of a leech from the South American *Helobdella triserialis* complex. – *Conservation Genetics*, 6: 467–472.
- UTEVSKY, S. & MAZEPA, G. (2005): A record of the South American leech *Helobdella triserialis* (Hirudinea: Glossiphoniidae) from an aquarium in Kharkiv, Ukraine. – *Lauterbornia*, 52: 177–180.

Kristóf MÁLNÁS, Zoltán MÜLLER, Béla KISS

BioAqua Pro Ltd.

H-4032 DEBRECEN, Hungary

Soó Rezső u. 21.

E-mails: malnask@gmail.com, mullerz@bioaquapro.hu, bkiss@bioaquapro.hu

Krisztián KOVÁCS

Government Office of Győr-Moson-Sopron County, Department for Environmental Protection and Nature Conservation, Laboratory for Environmental Protection

H-9028 GYŐR, Hungary

Török Ignác u. 68.

E-mail: krik@freemail.hu

Márk FICSÓR

Government Office of Borsod-Abaúj-Zemplén County, Department for Environmental Protection and Nature Conservation, Laboratory for Environmental Protection

H-3530 MISKOLC, Hungary

Mindszent tér 4.

E-mail: ficsor.mark@emikofe.kvvm.hu

Péter JUHÁSZ, Péter OLAJOS

Hortobágy National Park Directorate

H-4032 DEBRECEN, Hungary

Sumen u. 2.

E-mails: juhasz.peter@hnp.hu, olaj@hnp.hu