

## First report of potential pathogenic fungus *Papiliotrema aurea* in *Glossiphonia complanata*: implications for leech-mediated fungal transmission

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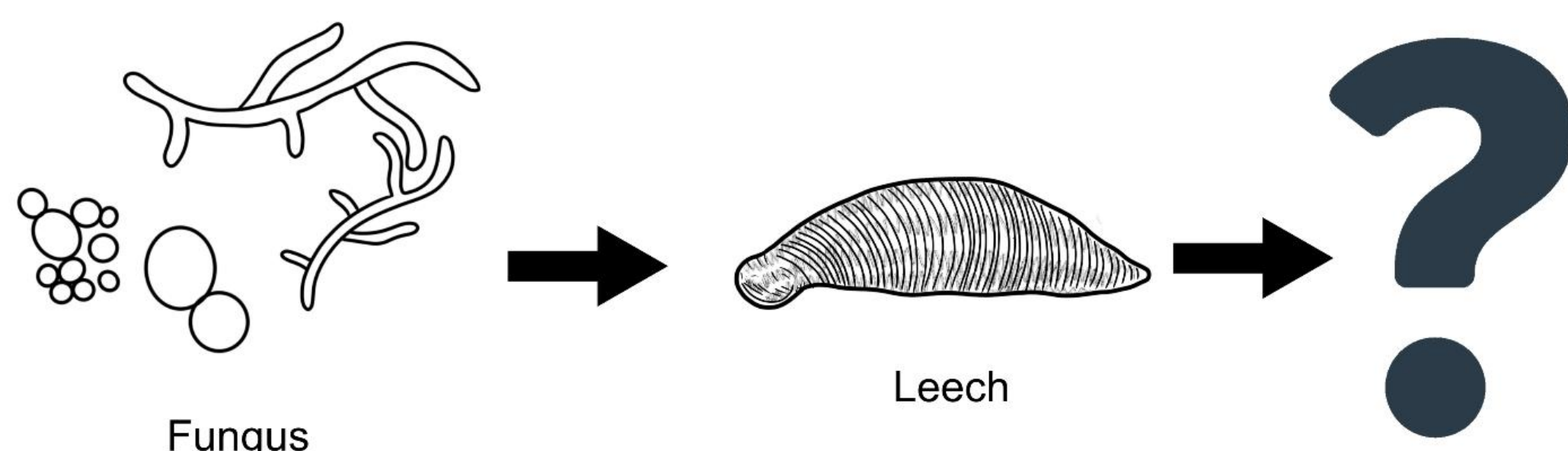
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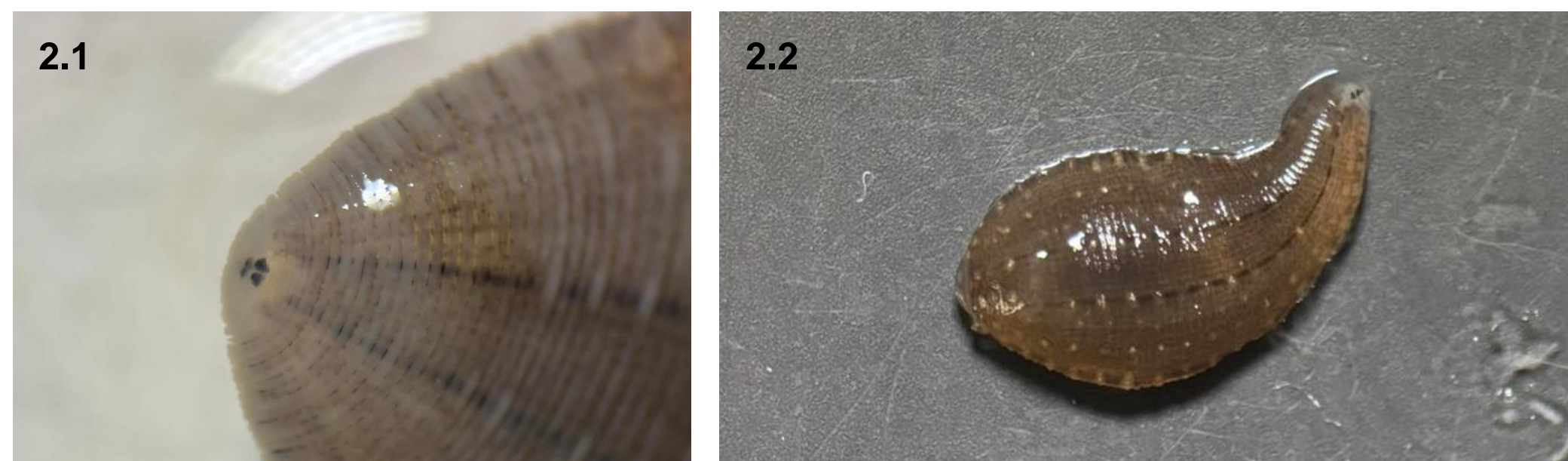
### INTRODUCTION & AIM

Leeches (*Hirudinea*) are a diverse group of annelids, most commonly associated with aquatic ecosystems, where they are involved in numerous interactions with other organisms. Depending on their feeding strategy, leeches can be classified as either parasites or predators.



**Fig. 1.** Leech as a potential vector of pathogenic fungi.

Due to dispersal capabilities and broad range of hosts, leeches may act as potential vectors for pathogens, including pathogenic fungi.

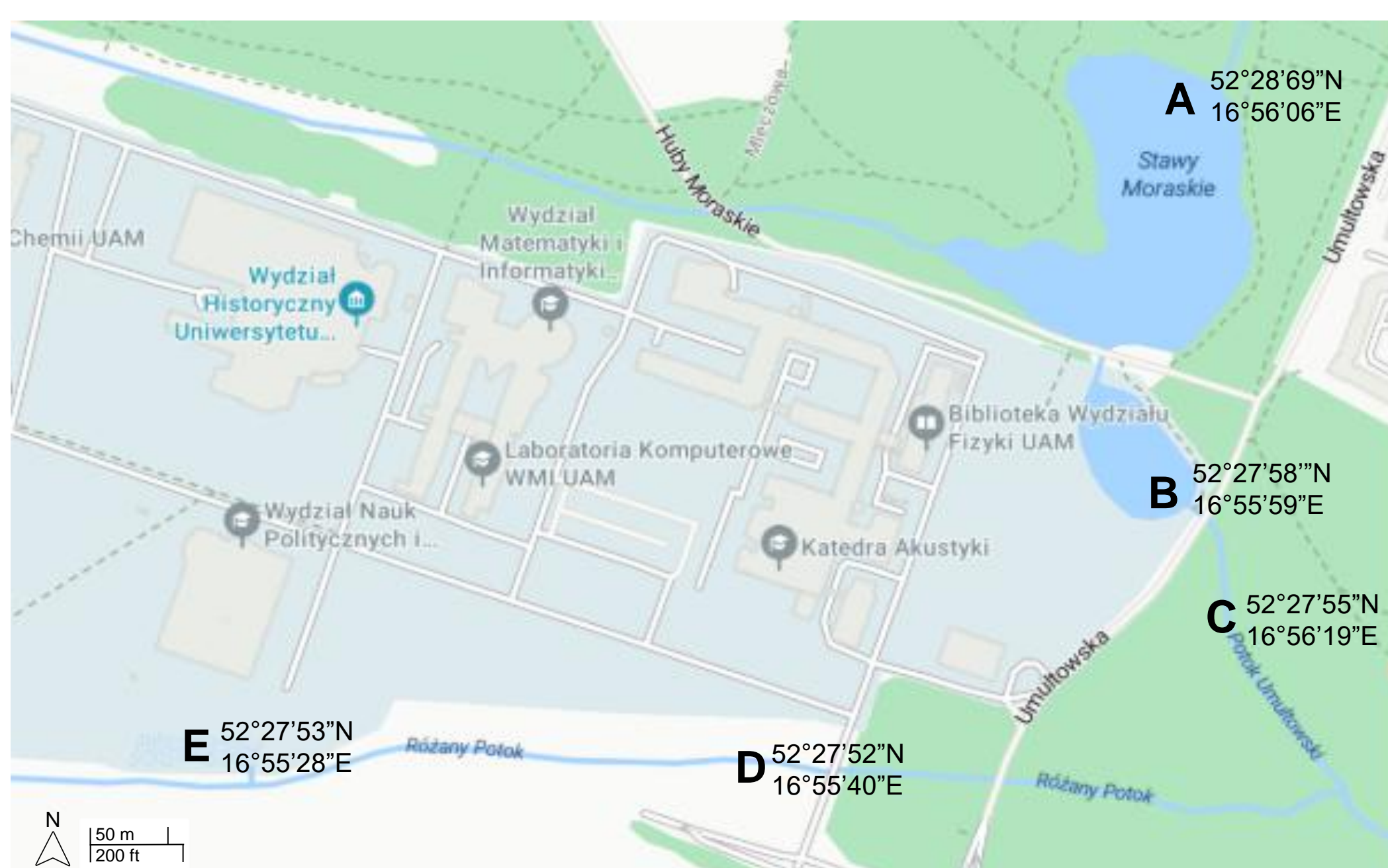


**Fig. 2.** Anterior part of the body (2.1) and dorsal side (2.2) of *Glossiphonia complanata* observed under a stereomicroscope.

The aim of the study was to analyze the fungal species diversity inhabiting both the external surfaces and internal tissues of leeches from the *Glossiphoniidae* family. This group was selected due to the variety of feeding strategies among its representatives, which include: (1) small predatory leeches preying on small invertebrates; (2) medium-sized parasitic leeches feeding on mollusks, primarily snails; (3) medium-sized leeches feeding on the blood of small vertebrates (fish, amphibians, reptiles); and (4) large parasitic leeches specialized in feeding on waterfowl, mostly ducks.

### METHOD

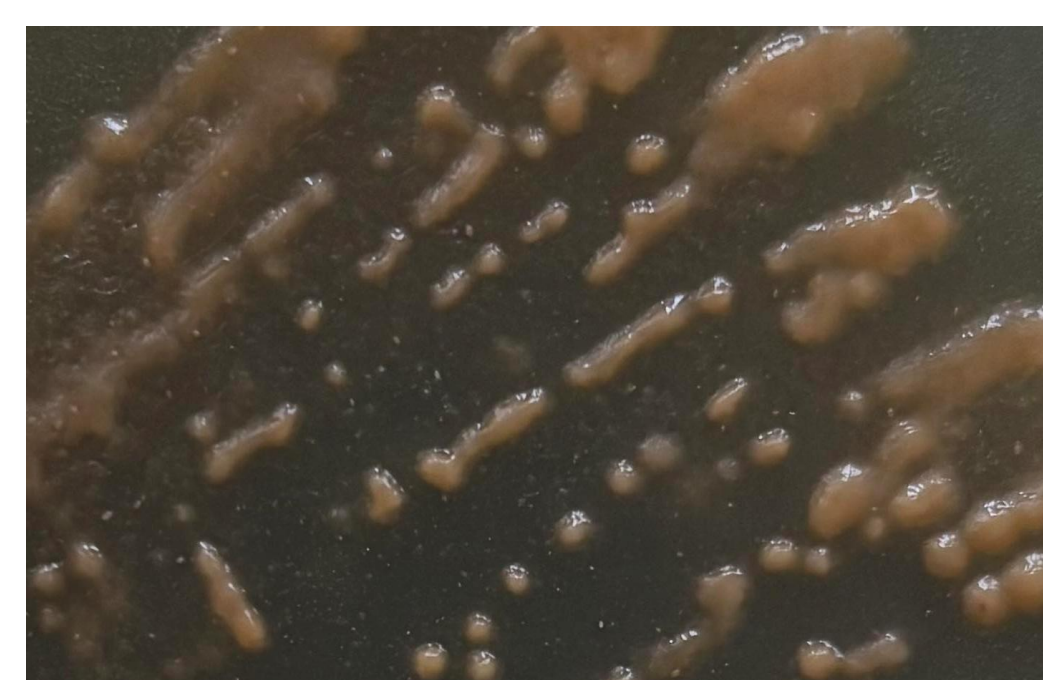
Leeches were collected from five water bodies located on the Morasko campus of Adam Mickiewicz University in Poznań [Fig. 3]. The dormant leeches were surface sterilized with 75% ethanol, divided into smaller fragments, and placed on PDA medium. The Petri dishes were incubated at 25°C, and any emerging fungal colonies were regularly transferred to fresh PDA medium. DNA was isolated from all colonies, and the ITS region was amplified using PCR. The amplification products were sent for sequencing. The obtained sequences were compared with sequences available in the EMBL and NCBI databases using the BLAST program. Additional morphological analyses were performed, and the isolated fungi were identified.



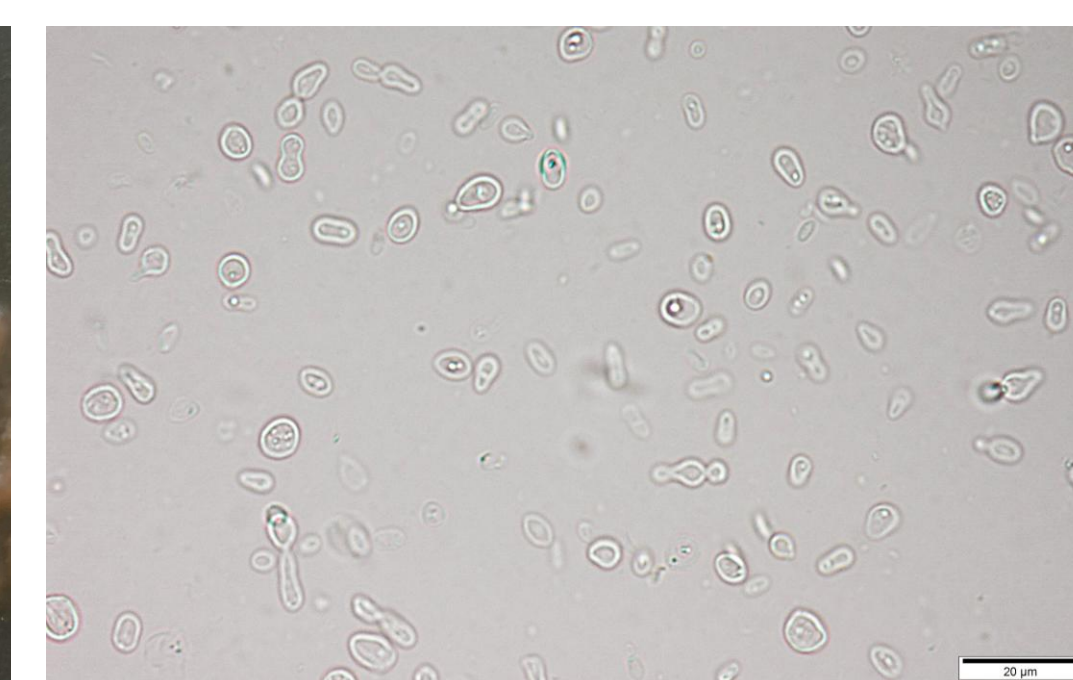
**Fig. 3.** Collection sites of leeches on the Morasko Campus, with five water bodies marked: (A) Staw Północny, (B) Staw Południowy, (C) part of the stream „Umultowski Potok”, (D) part of the stream „Różany Potok”, (E) retention reservoir behind the Collegium Politicum building, UAM.

### RESULTS & DISCUSSION

A total of 16 leeches were collected, with fungal growth observed in only 6 of them on a PDA medium prepared from fragmented specimens. [In addition to common environmental fungi such as *Cladosporium*, *Papiliotrema aurea* was isolated from the leech *Glossiphonia complanata* Fig. 4; Fig. 5]. This leech was collected from reservoir D, which is a 6,6 km long stream.



**Fig. 4.** Colonies of *P. aurea* on PDA medium under a stereomicroscope



**Fig. 5.** *P. aurea* cells under a light microscope

*Papiliotrema aurea*, previously isolated from the Chinese mitten crab (*Eriocheir sinensis*), demonstrates a significant capacity for colonizing a range of host organisms. This adaptability to diverse microenvironments suggests a need for further research, as limited data are available on its potential pathogenicity. Given its ability to colonize different hosts, *P. aurea* could pose an underestimated risk in various ecological contexts.

### CONCLUSION

- Leeches feed on various groups of organisms, demonstrating a high diversity in their choice of hosts.
- *Papiliotrema aurea*, previously considered capable of colonizing other species, has been isolated from the internal tissues of *Glossiphonia complanata*.
- Leeches may act as vectors in the spread of fungal diseases, which increases their potential significance in ecosystems and the health of other organisms.