

Review of: "A living bdelloid rotifer from 24,000-year-old Arctic permafrost"

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Bdelloid rotifers are amazing and mysterious organisms and known as scandalous creature thriving for millions years without sex. They are able to survive extremely harsh conditions including drought and freezing. Even knowing their resilience one can be totally astonished reading a last paper by Lyubov Shmakova, Stas Malavin, Nataliia Iakovenko, Tatiana Vishnivetskaya, Daniel Shain, Michael Plewka and Elizaveta Rivkina entitled: "A living bdelloid rotifer from 24,000-year-old Arctic permafrost". As it is hard to believe that multicellular organism is able to survive being frozen for so many years and then "wake up" without any damages, ready to grow and proliferate, my first doubts concerned possibility of the organisms being contamination of the samples. However, the researchers collected samples at a depth of 3.5m below the ground level extracting core and sampling in a way preventing contamination from upper layer. Basing on parallel studies from other laboratories they excluded possibility of small particles movement through ice or ice-cemented ground, so they assumed that isolated rotifers were trapped in permafrost at the same time as the radiocarbon-dated organic particles. Analysis conducted in Accelerator Mass Spectrometry dated sample as 23,960 – 24,485 years old. Knowing how easy it is "to get" Bdelloids from surrounding environment I felt relieved reading that in every step of sampling, transport of samples, culture and analyses researchers have made every effort to prevent contamination. Clear results of their effort are video sequence and picture of strain SCL-15-7 of *Adineta* sp. isolated from melted permafrost samples. Researchers not only isolated few strains of Bdelloid rotifers, cultured them, made pictures and videos, confirmed morphological determination with genetic analyses but they also tested their ability to survive freezing in the laboratory conditions. The phylogenetic position of ancient rotifer was also determined using modern molecular methods.

To confirm that isolated rotifers indeed originated from permafrost core, researchers searched for their sequences in a metagenome obtained from the same core sample and as authors stated "these results strongly suggest that the isolate originated from the permafrost layer and not sample contamination". The paper is short, well written and easy to follow. Most of the work done "behind the curtain" is described in every detail in the supplementary material giving the impression of solid, multidisciplinary work. I hope that this amazing discovery will encourage followers to use similar methods to disentangle mysteries of surrounding frozen world. Presented paper is valuable not only in the context of evolutionary biology but as a way to predict an environmental consequences of melting permafrost as a response to global

warming.

My concern is that following links in the supplementary material do not work:

github.com/stasmalavin/

[sleepingbeauty/blob/main/Borehole_AL3-15_isolated_organisms.csv](https://sleepingbeauty.blob/main/Borehole_AL3-15_isolated_organisms.csv)

github.com/stasmalavin/

[sleepingbeauty/blob/main/GMYC-analysis.R](https://sleepingbeauty.blob/main/GMYC-analysis.R)

github.com/stasmalavin/

[sleepingbeauty/blob/main/Primers-Actin.fasta](https://sleepingbeauty.blob/main/Primers-Actin.fasta)

ebi.ac.uk/Tools/msa/mview

github.com/stasmalavin/

[sleepingbeauty/blob/main/Quality-correction-alignment.fasta](https://sleepingbeauty.blob/main/Quality-correction-alignment.fasta)

github.com/stasmalavin/

[sleepingbeauty/blob/main/Actin-alignment-with-Sanger.fasta](https://sleepingbeauty.blob/main/Actin-alignment-with-Sanger.fasta)

github.com/stasmalavin/

[sleepingbeauty/blob/main/Actin-alignment-without-Sanger.fasta](https://sleepingbeauty.blob/main/Actin-alignment-without-Sanger.fasta)

github.com/stas-malavin/sleepingbeauty/blob/main/Actin-tree-without-Sanger.nwk