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Conference Abstract

New kid on the block: Research of alpine-lake macroinvertebrates in the Tatra Mountains enhanced by DNA barcoding and metabarcoding

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Abstract

State of ecosystems and biodiversity protection are becoming the key interests for modern society due to climate change and negative human impacts (Leese 2018). Environmental changes in freshwaters are indicated also by benthic communities, especially in sensitive ecosystems like alpine lakes (Fjellheim 2009). Moreover, remoteness and isolation of alpine lakes make them a source of biodiversity, which is worth conserving (Hamerlík 2014). A promising tool for efficient large-scale monitoring of aquatic communities is DNA metabarcoding (Leese 2018). In this study, we applied metabarcoding to analyse macrozoobenthos of 12 lakes in the Tatra Mountains, using benthic bulk samples and eDNA filtered from water (Fig. 1).
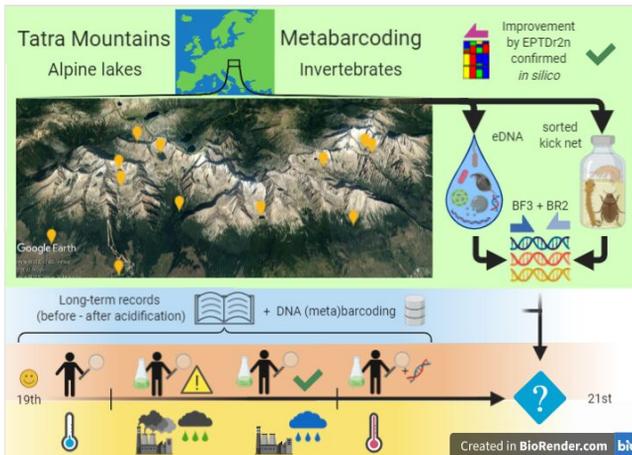
Figure 1. doi

Graphical abstract visualising study setup

In compliance with recent publications, eDNA amplified with BF3/BR2 primers resulted in high percentage of non-invertebrate reads (Leese 2021). Based on *in silico* tests with the obtained sequences, we confirm that the recently developed EPTDr2n primer enables minimizing non-target amplification even with eDNA filtered from alpine-lake water (Elbrecht and Leese 2017). This ability is facilitated by 3' end of the primer and we observed the two important mismatches in non-target sequences from our study (Leese 2021). Thus, our future analyses of eDNA (and bulk-sample fixative) will benefit from the new primer.

Concerning bulk samples, a wide range of invertebrate taxa was assigned to the OTUs and they showed good congruence with previous studies using morphological determination (e.g. Krno 2006). Certain differences with (and among) the previous records per lake were observed, which could suggest ecological changes, but at the moment the influence of sampling error cannot be excluded. In eDNA, several taxa were congruent with the previous records, but their amount and read abundance was considerably lower due to non-target amplification. Apart from that, filling gaps in barcoding databases remains one of our priorities, as identification to species or genus level was not yet possible for some invertebrate OTUs.

In addition, we subjected the NGS data to denoising and abundance-filtering in order to explore haplotype-level diversity (Andújar 2021). Although more comprehensive conclusions will be possible only after obtaining data from more lakes and years, already the two metabarcoding experiments presented here enabled us to efficiently detect within-species genetic diversity and identify a large variety of taxa, including groups that would otherwise be omitted or very challenging to identify. This underlines the potential of DNA methods to provide valuable ecological and biodiversity data across the tree of life for modern biomonitoring. This study was realized with support from VEGA 2/0030/17 and VEGA 2/0084/21.

Keywords

DNA, metabarcoding, invertebrates, Tatra Mountains, alpine lakes, biodiversity

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