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Global worming: an attempt to reconstruct earthworm paleohistory with eDNA

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Abstract
Earthworms are soft tissue organisms that rarely leave fossils that can be used to identify species. Absence of fossils makes the natural history of earthworm species in post-glacial landscapes of Fennoscandia largely unknown. Analyses of environmental DNA (eDNA) preserved in natural archives such as lake sediments and buried soil layers (paleosols) may offer an opportunity to assess the composition of past earthworm communities. In this thesis, I explore the use of metabarcoding as an analytical method to detect DNA from earthworms that lived in past European environments. I aimed at extracting DNA from various forms of paleosols in Europe and lake sediments, but earthworm DNA is rare in these deposits and amplifying DNA from this group of soil fauna was largely unsuccessful. However, during the scientific progression of my work, I discovered that metabarcoding-based studies are sensitive to 'tag jumping', which is a process where sample specific labels (tags) added to sequences for identification of individual samples 'jump', resulting in cross-talk between samples. My results suggest that tag jumping i) is mediated by the formation of heteroduplexes (DNA with two strands from different samples), ii) affects interpretations of eDNA studies by adding species to samples where they were not originally present, and iii) makes eDNA assemblages more similar. Importantly, my results also highlight that metabarcoding can generate powerful and trustworthy reconstructions of past environments if conducted with protocols that remove the influence of tag jumps. Reconstructions of terrestrial organisms from eDNA in sediments are also enhanced by erosion events that amplify DNA signals of land-living organisms. I conclude that earthworm DNA is difficult to detect in natural archives using current metabarcoding techniques and that tag jumping, a problem rarely discussed in metabarcoding studies, constitutes a concern in parity with direct sample contamination.

Keywords
Invasive, non-native, earthworm, paleohistory, migration, dispersion, sediment, eDNA, artifact, methods

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