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Environmental DNA metabarcoding tools for freshwater conservation: parallel characterisation of vertebrate and invertebrate communities.

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Surface freshwater is sporadic in its occurrence throughout much of semi-arid and arid Australia and where it does occur, it is often ephemeral. This is the case for freshwater granite rock-holes throughout remote southern Australia. These ecosystems act as a source of freshwater for a vertebrate community as well as habitat for aquatic invertebrates that display behavioural and physiological adaptations to the ephemerality of these systems. Climate change and the presence of invasive species are ongoing threats to this ecosystem, meaning it is likely to experience disruptions to historical hydrological patterns and potential declines. However, despite the ecological significance of the system and the likely threats, these rock holes are poorly understood. To establish baseline ecological information, we documented vertebrate and invertebrate species richness and variability at seven rock-holes in the Gawler bioregion of South Australia using environmental DNA (eDNA) metabarcoding methods. We recorded 19 vertebrate species and 22 orders of invertebrates, representing 45 families; finer taxonomic resolution was limited by a lack of robust molecular reference databases. Our findings demonstrate the importance of rock-holes as a water source for a wide range of endemic taxa and highlight the need for improved barcode reference libraries and associated metadata for accurate assignment of taxonomy to eDNA metabarcoding data in freshwater systems.

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