Evaluation of eDNA as a rapid biodiversity monitoring tool for great crested newt *(Triturus cristatus)*: a Scottish case study.

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Abstract:
Environmental DNA (eDNA) is DNA extracted from environmental samples and amplified using Polymerase Chain Reaction (PCR). It presents a non-invasive, low cost alternative to traditional monitoring for great crested newt *(Triturus cristatus)* and has been implemented in the UK and Europe. eDNA analysis was compared with torchlight survey for monitoring species presence of *T. cristatus* in 24 ponds at Gartcosh Nature Reserve in North Lanarkshire, Scotland. Primer sets designed for PCR and quantitative PCR (qPCR) were compared for amplification of *T. cristatus* DNA extracted from water samples using PCR. Torchlight survey recorded breeding adult counts, which were compared to data from 2006 – 2013 to assess current population status. Population viability and extinction risk were then investigated with a Population Viability Analysis (PVA). Torchlight survey was highly effective (83.7% - 100%) for detection of *T. cristatus* but results from eDNA analysis were affected by contamination. The qPCR primers appeared to amplify *T. cristatus* eDNA but results could not be verified by sequencing, whereas the PCR primers did not amplify eDNA. The Gartcosh population has increased since 2006 but the PVA indicated high extinction risk without continued conservation. This study was unable to recommend the eDNA method for future monitoring of *T. cristatus* but logistical advantages of the method were confirmed. eDNA analysis should not be used alone in Scotland until results are supported by sequencing of amplified products and the possibility of false negatives/positives is removed. Furthermore, traditional methods are still required for estimation of population size and identification of different life stages of *T. cristatus*. Resolution of the issues described in this article will improve reliability of the eDNA method and enhance monitoring for *T. cristatus* in Scotland.