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Conference Abstract

To blend or not to blend? The role of morphological traits for the detection of marine macrobenthos in bulk DNA and eDNA from the ethanol preservative

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Abstract

The impact of methodological choices on the reliability and reproducibility of DNA metabarcoding need to be well understood to allow successful implementation in routine monitoring frameworks. For macrobenthos communities, the metabarcoding protocol focuses on a fragment of the mitochondrial COI gene and depending on the primer set used for amplification of COI, different taxa can be detected. To identify the primer set that allows the best diversity estimates for macrobenthos in the North Sea region, we sampled four distinct and well characterised communities and identified macrobenthos using traditional morpho-taxonomy before molecular processing. Of the five primer sets tested, the Leray primer set yielded the highest number of non-chimeric reads, detected the highest number of macrobenthos species and best recovered beta diversity patterns. Despite the availability of a nearly complete reference database, 19 out of the 59 morphological species were not picked up with DNA metabarcoding. Next to primer choice, the DNA source used in metabarcoding studies can affect whether or not a species is

detected. DNA can be extracted from bulk specimens or from the ethanol preservative in which the macrobenthos sample was preserved. The latter DNA source would greatly speed up processing time of samples in the laboratory. We therefore compared species detection in bulk DNA and eDNA from the ethanol preservative from the four macrobenthos communities in the North Sea. Our results show that community composition differed significantly between bulk DNA and eDNA samples, but both sample types are able to differentiate the four macrobenthos communities from the North Sea. Of the 49 species that are detected in both sample types, 27 are also found in the morphological dataset. The 14 species that are exclusively detected in the ethanol preservative are mainly pelagic species. In view of the low read numbers allocated to these species (at most 153 reads) they most likely represent “contaminant” DNA molecules that are attached to the specimens or the organic debris. To better understand the different results between bulk DNA and eDNA from the ethanol preservative, we investigated the importance of four categorical traits in explaining the probability of detecting a species in the two sample types: body, larval stage (benthic or pelagic), longevity and body skeleton (chitin, $CaCO_3$ or soft tissue). A generalized linear mixed effects model approach shows that the probability of detecting a species in the eDNA from the ethanol preservative is significantly lower than for bulk DNA for macrobenthos species having small to medium body size and for species having chitine or $CaCO_3$ in their skeleton. In contrast, detection in the bulk DNA samples is not affected by the investigated traits. Although the ethanol preservative can be used to characterize beta diversity patterns, our results show that monitoring of macrobenthos species will be most robust when using bulk DNA as template for metabarcoding.

Keywords

DNA metabarcoding, ethanol preservative, reference database, morphology, eDNA, bulk DNA

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