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TITLE: Next Generation Sequencing Reveals the Hidden Diversity of Zooplankton Assemblages

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ABSTRACT:

Background Zooplankton play an important role in our oceans, in biogeochemical cycling and providing a food source for commercially important fish larvae. However, difficulties in correctly identifying zooplankton hinder our understanding of their roles in marine ecosystem functioning, and can prevent detection of long term changes in their community structure. The advent of massively parallel next generation sequencing technology allows DNA sequence data to be recovered directly from whole community samples. Here we assess the ability of such sequencing to quantify richness and diversity of a mixed zooplankton assemblage from a productive time series site in the Western English Channel. Methodology/Principle Findings Plankton net hauls (200 µm) were taken at the Western Channel Observatory station L4 in September 2010 and January 2011. These samples were analysed by microscopy and metagenetic analysis of the 18S nuclear small subunit ribosomal RNA gene using the 454 pyrosequencing platform. Following quality control a total of 419,041 sequences were obtained for all samples. The sequences clustered into 205 operational taxonomic units using a 97% similarity cut-off. Allocation of taxonomy by comparison with the National Centre for Biotechnology Information database identified 135 OTUs to species level, 11 to genus level and 1 to order, <2.5% of sequences were classified as unknowns. By comparison a skilled microscopic analyst was able to routinely enumerate only 58 taxonomic groups. Conclusions Metagenetics reveals a previously hidden taxonomic richness, especially for Copepoda and hard-to-identify meroplankton such as Bivalvia, Gastropoda and Polychaeta. It also reveals rare species and parasites. We conclude that Next Generation Sequencing of 18S amplicons is a powerful tool for elucidating the true diversity and species richness of zooplankton communities. While this approach allows for broad diversity assessments of plankton it may become increasingly attractive in future if sequence reference libraries of accurately identified individuals are better populated.

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