EVALUATING DNA-BARCODE BASED MONITORING OF BIODIVERSITY USING NEXT-GEN SEQUENCING

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ABSTRACT

A pilot study was performed to assess DNA-barcode based monitoring using environmental DNA to mirror biodiversity of two chosen habitats, a coastal heath and a Sitka spruce plantation, in Østerild in north-western Denmark. We evaluated the practicability of eDNA metabarcoding using current high-throughput NGS technology. The plantation was planted in the late 1950's on coastal heath and it was hypothesised that the species-composition of the bottom of soil cores collected in the Sitka-plantation would mirror the composition or represents some of the species composition found in the present coastal heath. Four plots, two in each habitat were chosen based on coherence between information from vegetation analysis and species-assessment based on arthropods trapped in pitfall traps collected during another project. To test whether metabarcoding identification provides meaningful species/genus lists compared to the ones obtained from the conventional inventory, soil cores from the plots were analysed. eDNA from only 250 mg soil was extracted from eight samples (four plots x top/bottom) using two different methods. Two separate eDNA analyses were performed using different NGS-techniques: The first approach targets four different organism groups (plants, insects, collembolans, bacteria) using PCR amplification of *trn*, COI, 18S and 16S markers followed by sequencing (Ion Torrent/PGM). By comparison against a nucleotide database (NCBI-nt) reads were annotated to the highest resolution – species for some, only genus or even family for others. In the second approach random sequences of the genomic eDNA were generated (Illumina HiSeq2000) and compared to a non-redundant protein database (NCBI-nr) and bacterial genomes. Data from both NGS-approaches are in the process of being analysed aiming (1) to identify differences in the species-composition between the two habitats comparing different NGS-methods with conventional methods and (2) to evaluate if the species composition of the bottom soil cores in the Sitka-plantation represents a sub-set of the composition characterising the coastal heath.