

Soil amoebae: Identification of new species and evaluation of the community structure

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Protozoa and especially amoebae are the major consumers of bacterial production in soil, forming the base of the heterotrophic eukaryotic food web that channels the energy flow via bacteria to higher trophic levels in the soil food web (i.e. the bacterial energy channel).

As part of the EU-project EcoFINDERS, we have been combining microscopic and molecular high-throughput sequencing techniques and determined the diversity of amoebae in different soils across Europe and in high altitude soils in Tibet. This information is essential to identify the functions of the major amoeboid taxa in soils.

Cultivation and subsequent morphological and molecular description of amoeba strains from diverse soils indicated a high level of species diversity and probable existence of new lineages corresponding to genera and even higher taxa. A detailed analysis of soils from high altitudes in Tibet revealed 25 distinct sequences within the supergroup Amoebozoa belonging to 15 species which could be distinguished morphologically. Combined morphological and phylogenetic information revealed that at least 11 clones represent previously non-described species. We found a high diversity of taxa and even new genera especially in the class Varioseae. Similarly, amoebae from the class Heterolobosea within the supergroup Excavata were species-rich. We could describe several new species within the genus *Allovalkampa* and even a new genus currently only known by environmental sequences.

We also investigated the diversity of soil amoebae in a 454 high-throughput sequence study with newly designed *Acanthamoeba*-specific primers. Within this study, ten independent soil replicates from two land-use intensities were analysed which were located at six distinct sites across Europe. On average, we obtained 1000 sequences with a large diversity of different *Acanthamoeba*-specific sequences at each site. These included many currently unknown ones suggesting an enormous hidden diversity even within a comparably well-described genus of amoebae.