COMPARATIVE ANALYSIS OF 16S r-RNA GENES FROM PIORA DOLOMITE REVEALS A NEW GROUP OF ENDOLITHIC ARCHAEA

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The conditions in certain carbonate and sandstone rocks allow organisms, mostly microorganisms, not only to survive but to develop to well adapted endolithic communities. In this study, we focused on the presence of Archaea and discovered a new group which branches within the Crenarchaeota. The new 16S rRNA sequences cluster together and are clearly different from their closest relative *Cenarchaeum symbiosum*, a non-thermophilic, symbiotic Crenarchaeote from marine sponges. The evolutionary distance between *Cenarchaeum symbiosum* and the newly discovered cluster is more than 10%.

The results presented are part of an ongoing study on endolithic microbial communities in the Piora Dolomite. Piora Dolomite, which is found in several outcrops near Lago Cadagno, is a gypsum bearing granular magnesium-calcium carbonate. It contains brown to greenish-grey bio-layers, which penetrate 1 to 5 millimetres deep into the rock.

DNA was extracted and the DNA templates were amplified with archaeal, bacterial, universal and eukaryote specific primer pairs employing the Polymerase Chain Reaction (PCR). PCR products were cloned in order to separate the different 16S and 18S rRNA gene fragments. Restriction enzymes HaeIII and HinfI were used to analyze the fragment length polymorphism (RFLP) and clones with different RFLP patterns were selected for sequencing. Ten different archaeal and five eukaryote specific sequences were recognized. These were compared using cluster analysis employing the Phylip program and the results are presented in the form of a phylogenetic tree based on sequences present in the RDP 16S rRNA gene library.

The eukaryotic sequences which were found belong to a cluster of Bryophytes (mosses), closely related to *Leptobryum pyriforme*. The presence of these mosses on or in dolomite has not been known so far.

Our results show the presence of a number of Archaea in dolomite rock. Together with the bacterial and amoeba like sequences, which were found in earlier studies, they make up the fascinating community of rock inhabiting microorganisms. For the time being the nutrient sources in the dolomite rock, as well as the biochemical interactions between the members of the community remain questions to be resolved next.