

SHORTCOMINGS OF CULTURING BACTERIA FROM THE DEEP SEA ANOXIC BASINS (MEDITERRANEAN SEA)

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Abstract

Culturing of prokaryotes remains the ultimate goal for a detailed characterization of the metabolic pathways of microorganisms despite recent advances in genetic analysis of functional genes. We have isolated previously uncultured bacteria from four deep-sea hypersaline anoxic basins of the eastern Mediterranean Sea. Molecular analysis of isolated bacterial strains point towards a rich bacterial community present in this extreme environment.

Keywords: bacteria, culturing, deep sea, anoxic basins, Mediterranean Sea

Despite the introduction of culture-independent molecular screening techniques that allows microbiologists to examine a wide spectrum of microbial diversity, culturing techniques are still valuable and might reveal information which cannot be obtained by molecular techniques (1; 2; 3). The inability to culture bacteria has dampened efforts to study the links between bacterial taxonomic diversity and their functional role in the native environment (4; 5; 6; 7). Due to their non-cultivability, the metabolic potential of the highest number of described bacterial taxa and, therefore, their possible industrial application remains unknown. Studying the organisms in culture can not only provide new information about microbial evolution and ecology but may also yield a host of useful compounds, such as antibiotics or enzymes with unexpected properties.

Within the BIODEEP EU project (EVK3-2000-00042) we have managed to grow in the lab several strains of previously unculturable bacteria adapted to thrive at "extreme" conditions—an advance that may provide a new means of exploring the vast diversity of microbial species. For such an aim, four deep-sea hypersaline anoxic basins (DHABs) located in the Eastern Mediterranean Sea have been sampled at different layers. The DHABs are completely perennially hypersaline and oxygen-free environments that originate from ancient subterranean salt deposits (evaporites). Due to their physical and chemical features, the DHABs could easily represent prime regions for unraveling the extent of microbial diversity and for determining the lower limits of life-supporting environmental parameters.

About 250 bacteriological samples from the DHAB brines, interfaces and sediments have been processed with different parallel technologies and a special attention for reducing the possibility of contamination. The environmental conditions at the sampling site were used in media design (e.g. looking for ingredients needed for the bacteria to survive). About 500 bacterial strains have been isolated and identified by means of 16S rDNA-based molecular analysis. The results point towards a rich, mainly high-salt adapted prokaryotic diversity which may be used as baseline information for the assessment of microbial communities of other marine hypersaline anoxic environments. A statistic approach was used for comparing the used culture media on the basis of the selected taxonomical diversity and relative abundance. According to that, several media appeared to be equivalent to each other whereas few of them were unique. Repeated culturing of the same strain from more than one DHABs sample supported the hypothesis of the existence of DHABs adapted cultivable bacteria with similar metabolic patterns.

The tools described represent state-of-the-art technologies, which may be among those applied to monitoring life forms in DHABs-like terrestrial (and extraterrestrial?) environments.

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