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# Preservation of microbial consortia: impact of storage at 4°C and -80°C on an As(III)-oxidizing community

Caroline MICHEL, Fabienne BATTAGLIA-BRUNET, Catherine JOULIAN

*BRGM, DEPA/GME (Geomicrobiologie et Monitoring Environnemental), 3 avenue Claude Guillemin, BP 36000, 45060 Orléans cedex 2 (France)*

The use of mixed microbial communities in biotechnologies offers many advantages compared to single/pure strains (Brenner et al., 2008):

- They can face environmental changes in an easier manner than monocultures can do.
- They can perform complicated tasks/functions such as that requiring multiple steps, which can be difficult or even impossible for many individual strains.
- Microbial consortia represent a source of genes and functions, as well as resources for future and novel biotechnological applications as they involve non-cultivable and/or uncharacterized microorganisms.
- Microbial communities are naturally mixed in the environment and consortia are thus more representative of the “real” life.

However, the preservation, especially over the long-term, of stable microbial consortia is still underlined as a challenge. It has become a necessity to safeguard the accessibility to microorganisms already recognized to have interesting features and to be a pool of microbial resources. Several factors are known to be critical for a good storage, such as cell size and type, temperature, amount of cells, physiological state, the use of a cryoprotectant for cryoconservation and lyoprotectants for lyophilisation, the period of time of cryoprotectant/cells contact before cooling, the rates of cooling and thawing... These parameters have been identified as critical for the storage of pure microbial strains but little is known on how to preserve microbial consortia.

Here, the impact of two ways of storage widely used in laboratory, cryoconservation at -80°C in 20% glycerol and 4°C storage, was studied on a microbial community selected for its ability to oxidize As(III) into As(V), an important function in the frame of As bioremediation purposes. The community was characterized before and after storage, over a period of up to 12 months, focusing on the following parameters: viability (Live and Dead labelling approach), cell growth (microscopic counts), activity (As(III) oxidation and carbon sources utilization Biolog profile), and diversity fingerprints. Results showed no impact of both ways of storage on As(III) oxidation ability and cell growth, but storage led to an impact on cells viability. An increase of biodiversity after storage at -80°C was observed, probably due to the presence of glycerol that may have allowed the growth of strains that were in minority before storage. On the opposite, storage at -4°C tended to decrease biodiversity. Results also showed an evolution in carbon source utilization that varied according to the temperature and the time of storage. In conclusion, the function of interest, As(III) oxidation, was well preserved whatever storage temperature and duration, but microbial diversity and viability were impacted, even during short term storage.

*Brenner K, You L, Arnold F (2008) Engineering microbial consortia: a new frontier in synthetic biology. Trends Biotechnol 26: 483-489.*

Key words: consortium preservation, mixed microbial community, arsenic bio-oxidation, activity, diversity.