

Deoxyribose Nucleic Acid (DNA) sequencing at the picogram level to investigate life on Mars and Earth

The DNA picogram detection level and the procedure presented here, may be of interest for the future **Mars sample Return (MSR)** program, and the life research and planetary protection studies that will be implemented on Earth.

Deoxyribose Nucleic Acid (DNA) is an incontrovertible **biosignature** whose sequencing aids in species identification, genome functionality, and evolutionary relationships. **To study life within the rocks of Earth and Mars**, we demonstrate, in an ISO5 clean room, a procedure based on **nanopore technology** that correctly identifies organisms at picogram levels of DNA without amplification. Our study with *E. coli* and *S. cerevisiae* DNA samples showed that MinION sequencer (Oxford Nanopore Technologies) can unequivocally detect and characterize microbes with as little as 2 pg of input with just 50 active nanopores. This result is an excellent advancement in sensitivity, immediately applicable to investigating low biomass samples. This value is also at the level of possible background contamination associated with the reagents and the environment. Cultivation of natural and heat-treated Martian analogue (MMS-2) regolith samples, exposed to atmospheric water vapor or in increasing water concentrations, led to the extraction of 600–1000 pg of DNA from 500 mg of soil.

Applying the low detectability technology enabled through MinION sequencer for a natural low biomass setting, we characterized the dry MMS-2 and found few soil-related organisms and airborne contaminants.

MSR Artistic representation. Image Credit: M.-P. Zorzano/ P. Blázquez