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Sensitive life detection: extraction of nucleic acids sorbing to Mars analogue minerals

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Abstract

The main goal of space missions to Mars is to find irrefutable proof of life. Consequently, development, evaluation and optimization of sensitive extraction and detection methods for biomarkers are of extreme importance. Our aim consisted in the optimization of sensitive extraction techniques for molecules storing hereditary information (nucleic acids such as DNA), since these are common in life forms. However, adsorption of nucleic acids to mineral matrixes and soils can generate low extraction yields. Therefore, a second aim was to determine adsorption and identify 'problematic' Mars analogue minerals. In addition, the development of a method for quantification of DNA recovery by the use of an internal control was proved to be essential, since sensitive extraction needs information on recovery.

1. Introduction

Extracting biomarkers from Martian analogue samples is challenging due to a wide range of physical and chemical conditions, and the likely low abundance (if at all) of life. Samples of interest may include sedimentary and brine deposits, clay-rich soils, and rocky matrixes such as volcanic and other variety of rock outcrops. Mars analogue minerals comprise silicates (olivine, pyroxene and plagioclase), evaporites (sulfates like gypsum and jarosite), iron oxides (haematite and magnetite), iron oxyhydroxides (goethite and ferrihydrite), carbonates, and phyllosilicates (clay minerals montmorillonite and nontronite). Some soils and particularly clay minerals are known to strongly adsorb DNA. In addition, previous experiments have shown that some Mars analogue soils adsorb or degrade DNA [1]. We aimed to quantify the nucleic acid adsorbing capacities of minerals and to optimize and quantify the recovery of nucleic acids from minerals that adsorb nucleic acids.

2. Results and Conclusions

Three different types of experiments were performed: the determination of DNA adsorption to Mars analogue minerals, the optimization of DNA extraction and the development of an internal control experiment by the use of a spike to quantify DNA recovery. Quantification was always done by qPCR. The mineral adsorption experiment revealed that clay minerals interfere with extraction by adsorbing DNA to over 99%. This result should be taken in consideration since clays are possible targets in the search for life. Therefore, it is important the optimization of extraction methods, mainly for clays. The experiments of optimization of extraction comprised three different approaches. The first approach consisted of a method to dissolve mineral matrixes, in particular by hydrofluoric acid (HF) and a combined acid and heat treatment. The aim of this experiment was the tentative separation of DNA and minerals by acid dissolution. These treatments destroyed DNA, although HF left the structure of cells intact so other biomarkers are likely preserved. Hot water extraction, the second approach, is an effective method for other biomarkers such as amino acids, which also degraded DNA. The third approach, the use of different extraction solutions to promote an improved extraction yield turned out to be the best. Higher DNA recoveries (up to 100 fold) were obtained with an elevated concentration of phosphate buffer. Yet, recoveries were about 10%. Therefore, we are developing a protocol using an internal spike to quantify DNA recovery.

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References

[1] Direito, S.O.L., Ehrenfreund, P., Marees, A., Staats, M., Foing, B., and Röling, W.F.M.: A wide variety of putative extremophiles and large beta-diversity at the Mars Desert Research Station (Utah), Int. J. Astrobiol., Vol. 10 (3), pp. 191–207, 2011.