

## Biosignatures of methanogenic archaea by Confocal Raman Microspectroscopy (CRM)

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Methanogenic archaea are anaerobic chemotrophic microorganisms that meet many of the metabolic and physiological requirements for survival on the martian subsurface. In particular, methanogens from Siberian permafrost are extremely resistant against different types of environmental stresses as well as simulated martian thermo-physical and subsurface conditions, making them promising model organisms for potential life on Mars. Raman spectroscopy is a vibrational spectroscopic technique that has shown a remarkable potential in microbial identification. It provides fingerprint-like information about the overall chemical composition of the samples and allows a nondestructive investigation. The biosignatures of *Methanosarcina soligelidi* SMA-21 were characterized by CRM during the growth phases at a single-cell level, which presented a high heterogeneity and diversity in the chemical composition of the cells and detectible subpopulation differences. This study also highlighted potential technical challenges concerning the Raman detection of methanogenic archaea (and other non-pigmented microorganisms) embedded on a mineral substrate. The biosignatures of permafrost and non-permafrost strains in the stationary phase of growth were also characterized by CRM. A cluster analysis of the spectra revealed that permafrost and non-permafrost strains have a different overall chemical composition, which has possible evolutionary implications.