Microbial diversity and biosignatures in extreme environments: the Alalobad geothermal field (Afar, Ethiopia) as a planetary analogue

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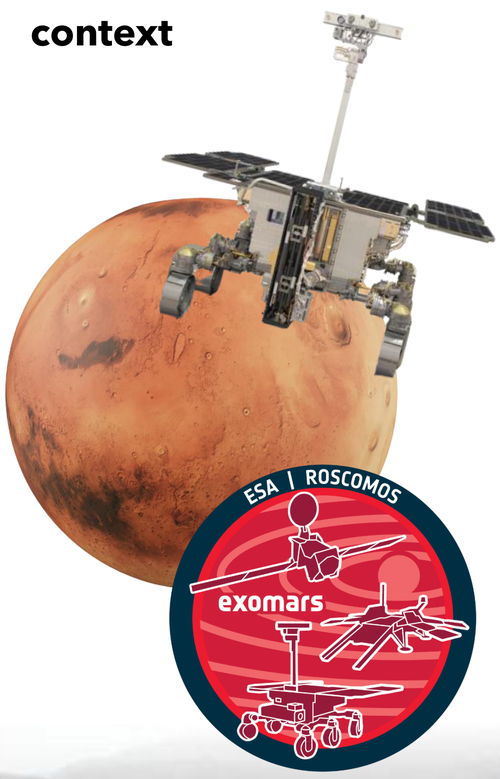
Astrobiological context



- Hydrothermal systems are ideal natural laboratories for studying **biosignatures**: they host steep physicochemical gradients, rapid mineral precipitation, and microbe–mineral interactions that enhance the preservation potential of biological signals.
- These settings closely resemble the conditions hypothesized for **early Earth** and **early Mars** and because of this strong geological and geochemical analogy, they serve as excellent **Planetary Field Analogues (PFAs)**.
- The upcoming **ESA ExoMars mission** reinforces the scientific interest in Mars as the primary target in the search for traces of past or present life.

Geological context

The Alalobad geothermal field is located on the western flank of the Tendaho Graben, within the actively extending Afar Depression. The site hosts a range of hydrothermal features, including **boiling pools** (up to ~99 °C), **fumaroles**, **mud pools**, and extensive **travertine and silica deposits** that reflect ongoing water–rock interaction.

Objectives

This study aims to:

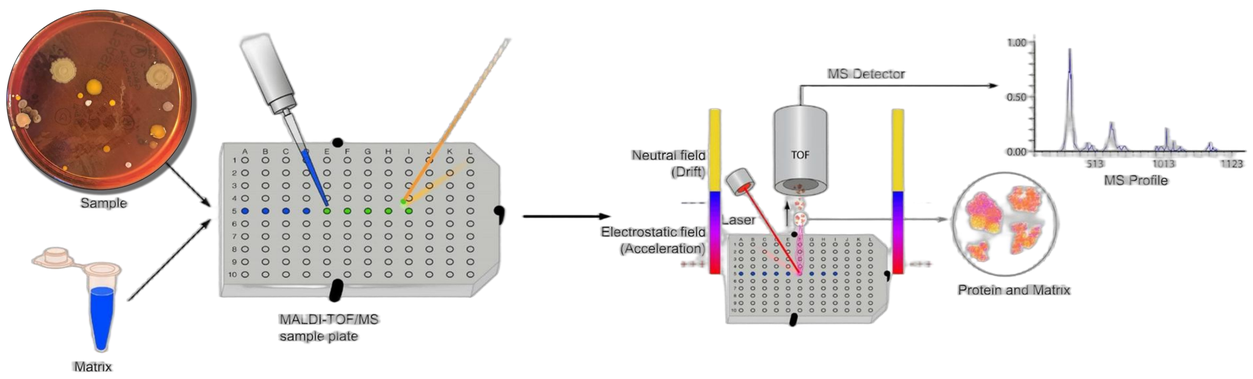
- Characterize the geological and microbial **habitats**
- Investigate the presence and **preservation potential** of biosignatures
- Evaluate the potential of Alalobad as analogue for early Earth and early Mars.

After Rizzello et al., 2021

Sampling strategy

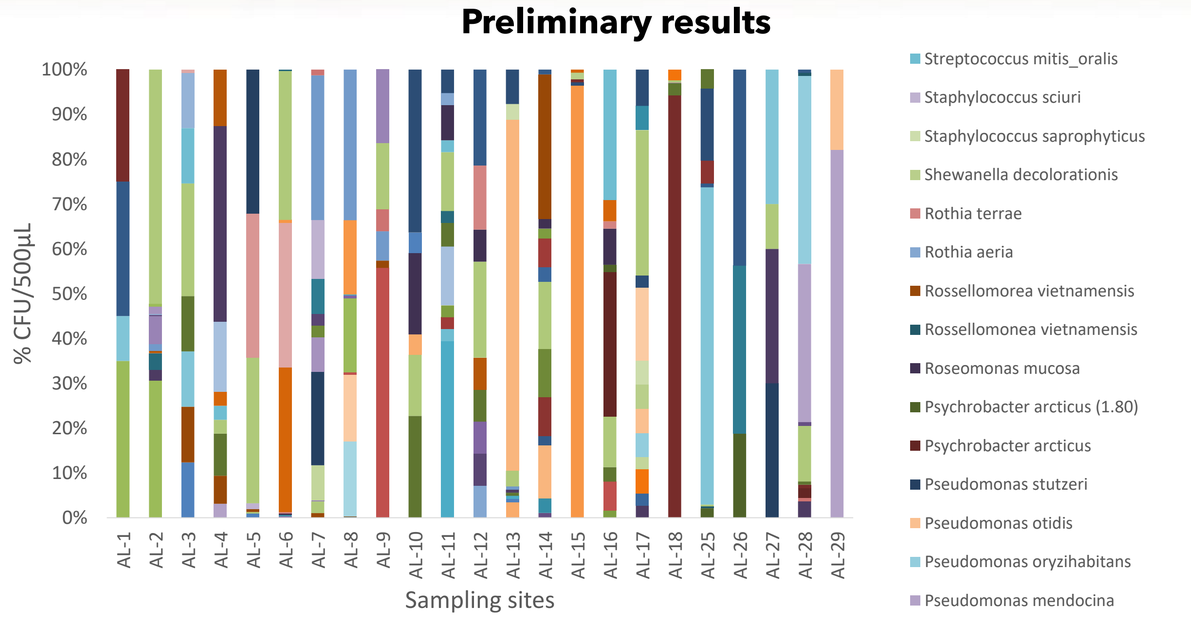
- **Sterile swabs** were used on microbial mats and pool margins to capture microbiological diversity across thermal and geochemical gradients.
- Additional samples included sediments, travertine, and silica-rich facies to characterize the geological context.
- In situ measurements of temperature, pH, conductivity, and site morphology were recorded for environmental framing.

From cultivation to taxonomic profiling



- The eluate from swabs was plated onto three different culture media: **Tryptic Soy Agar** (TSA), **Nutrient Agar** (NA), and **Blood Agar** (BA), then incubated at **20°C**, **30°C**, and **55°C** to maximise the recovery of the cultivable microbial fraction.
- After incubation, single colonies were selected from the plates and prepared for analysis by **Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry** (MALDI-TOF MS, Bruker Biotyper ®).
- It provides rapid, reproducible, and cost-efficient identification of environmental isolates.
- Individual colonies were transferred onto a MALDI plate and overlaid with matrix solution. Then the spectra were acquired and compared against reference databases to obtain score-based taxonomic identifications.

Preliminary results



The analysis of the cultivable fraction revealed a mix of well-characterized species and several non-identified, suggesting the presence of **potentially novel** or **underrepresented taxa** within the environmental databases.
The next steps include:

- shotgun metagenomic sequencing to characterise the non-culturable fraction
- geological and spectroscopic analyses (Raman, SEM–EDS) to investigate mineral–microbe relationships and biosignature preservation.

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