Origins: From the Protosun to the First Steps of Life
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Simulating putative Enceladus-like conditions: The possibility of biological methane production on Saturn's icy moon

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Abstract. In this study (Taubner *et al.* 2018), three different methanogenic archaea (*Methanothermococcus okinawensis*, *Methanothermobacter marburgensis*, and *Methanococcus villosus*) were tested for metabolic activities and growth under putative Enceladus-like conditions, including high pressure experiments and tests on the tolerance towards potential gaseous and liquid inhibitors detected in Enceladus' plume. In particular, *M. okinawensis*, an isolate from a deep marine trench (Takai *et al.* 2002), showed tolerance towards all of the added inhibitors and maintained methanogenesis even in the range of 10 to 50 bar. Further, we were able to show that H_2 production based on serpentinization may be sufficient to fuel such methanogenic life on Enceladus. The experiments revealed that methanogenesis could, in principle, be feasible under Enceladus-like conditions.

Keywords. astrobiology, planets and satellites: Enceladus, methods: laboratory

1. Introduction

Enceladus is one of the most remarkable objects in the Solar System. Besides its spectacular plume located at the South Polar Terrain, it is the origin of the plume particles which makes Enceladus one of the hot spots in Astrobiology these days: a global subsurface liquid water ocean (Thomas *et al.* 2016). The molecules detected in the plume (Waite *et al.* 2009, 2017), e.g. water (H_2O), carbon dioxide (CO_2), or methane (CH_4), in combination with the possibility of hydrothermal vents at the ocean floor (Hsu *et al.* 2015) led to the hypothesis that this icy moon might be habitable for life as we know it. The detection of molecular hydrogen (H_2) in the plume (Waite *et al.* 2017) corroborates this hypothesis, and serpentinization of the chondritic core of Enceladus is a potential source of H_2. The most promising area on Enceladus where life may exist is at the seafloor of the ice-covered ocean, where the pressure was estimated to be 35 to 80 bar (Hsu *et al.* 2015, Taubner *et al.* 2018).

Possible life forms on Enceladus must be chemoautotrophic, i.e. independent of the presence of organic matter, and would need to be anaerobic, i.e. independent of oxygen. Hydrogenotrophic methanogens, i.e. archaea that metabolize H_2 and CO_2 to produce CH_4 and H_2O, are among the organisms that meet these characteristics. In this study we raise the question if some of the CH_4 detected in the plume might originate from biological activity.

2. Experimental Design

The first step of the presented study (Taubner *et al.* 2018), was to screen the literature for suitable strains. As presented elsewhere (Hsu *et al.* 2015), the temperature close to the hydrothermal vents on Enceladus' ocean floor could be relatively high. Therefore and because of the higher growth rate, we decided to focus on (hyper-) thermophilic methanogens. After some preliminary tests, we decided to focus on the three methanogenic strains *Methanothermococcus okinawensis*, *Methanococcus villosus*, and *Methanothermobacter marburgensis*. The latter strain originates from a waste water treatment plant (Schönheit *et al.* 1980), whereas the other two strains were isolated from a shallow submarine hydrothermal system (*M. villosus*, Bellack *et al.* 2011) and a deep marine trench (*M. okinawensis*, Takai *et al.* 2002), respectively. To prioritise one of the above mentioned strains, we tested these organisms for tolerance towards potential gaseous and liquid inhibitors detected in Enceladus' plume (formaldehyde, ethene, methanol, and high ammonia levels). Only one strain, namely *M. okinawensis*, showed continuous and stable growth. We therefore selected this strain for the final high-pressure experiments. These experiments were performed in a high pressure system consisting of either a 0.7 or 2 L stirred Büchi reactor, a heating system, a vacuum pump, a transfer vessel (for inoculum and special media compounds), and a connection to a computer. The reactor can be controlled via remote-control and the appended gas mixture can be easily varied. We tested the methane production of *M. okinawensis* at different pressure values (ca. 10, ca. 25, and ca. 50 bar) with a Enceladus-like gas phase (ca. 30-40 Vol.-% N_2, ca. 40-55 Vol.-% H_2, ca. 5-7 Vol.-% CO_2, ca. 3-5 Vol.-% CO, ca. 3-6 Vol.-% C_2H_4) at 65°C. We were able to show that *M. okinawensis* was able to perform H_2/CO_2 conversion and CH_4 production under the above described Enceladus-like conditions. Even at the highest applied pressure of 50 bar, a CO_2 to CH_4 conversion of more than 70% was observed.

3. Serpentinization

In parallel, serpentinization-based H_2 production rates were modelled using the computer code PHREEQC (Parkhust *et al.* 1999) as part of this study to determine if serpentinization reactions on Enceladus can support a rate of H_2 production that is high enough to sustain autotrophic, hydrogenotrophic methanogenic life. Depending on the applied mineral content, a H_2 production rate of approximately 5 to 50 nmol g^{-1} L^{-1} d^{-1} was estimated (Taubner *et al.* 2018), which may be sufficient to serve as a substrate for biological CH_4 production on Enceladus.

4. Conclusion

This work (Taubner *et al.* 2018) presents the first systematic astrobiological study on methanogenic physiology under high pressure and may enable a new assessment of potential habitats in the Solar System, especially icy moons. This study does neither prove that there is life nor that there is an origin of life setting on Enceladus. However, it showed that methanogenesis could, in principle, be feasible under Enceladus-like conditions and that some of the CH_4 detected in Enceladus' plume could, in principle, be of biological origin. It also points out the importance of planetary protection regulations for future space missions to icy moons like Enceladus and leads to a rethinking of how we should define a habitable zone in a planetary system.

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Discussion

WANDEL: Could you comment on the difference between the two best solar system candidates, Europa and Enceladus?

TAUBNER: The most obvious difference is the larger volume of Europa and its subsurface ocean, which leads to a way-higher pressure at the seafloor of Europa compared to Enceladus.

WANDEL: In particular about the prospects for contact of the water body with the silicate bottom?

TAUBNER: According to current models, both Enceladus' and Europa's oceans are in direct contact with the silicate core.

FAN ZHANG: Have you tried to slowly introduce toxicity to the Earth organisms and see if they can evolve to tolerate the environment that didn't pass your selection criteria and get more options?

TAUBNER: This work is planned, not yet done.

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