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# Viability of Bacterial Spores Under Ocean Worlds Icy Surface Conditions

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## 1. Introduction

Ocean worlds like Europa or Enceladus are exciting targets to search for life in our Solar System, owing to the presence of liquid water and water-rock interactions. Micro-organisms formed there can potentially migrate to the surface via plumes, ice shell tectonics, or in chaos regions, and be accessible to an eventual landed mission. Laboratory studies are needed to better understand microbial degradation under the harsh conditions encountered on Ocean Worlds Icy surfaces to help in their detection. Furthermore, such studies are of high interest for planetary protection purposes in order to understand the risk of contamination by terrestrial life of outer Solar System landing targets.
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## 2. Methods

The goal of our work is to investigate micro-organism survival under the temperature, radiation, and in the icy medium found at the surface and near-surface of icy worlds. To accomplish this, we chose *Bacillus subtilis* spores, a dormant bacterial state as our model microorganism for Icy World endemic life, because of their known resilience under extreme conditions on Earth. In our laboratory, *B. subtilis* spores can be irradiated under high vacuum by UV photons using a mini-arc Ar lamp to mimic the Solar spectrum or an electron gun to simulate low pressure and energetic conditions on icy bodies. They can be cooled cryogenically in the 10 - 300 K range, and molecular films were deposited by introducing gaseous mixtures that condense on the cooled spore samples. Care was taken to deposit and irradiate bacterial spores in the sub-monolayer regime to prevent self-shielding (Noell et al., 2013). The spores viability was quantified by retrieving the microorganisms after irradiation, culturing them onto agar plates, and calculating the ratio of colony-forming units between irradiated and control samples.

## 3. Results and Discussion

Here we present viability fractions of *B. subtilis* spores irradiated by UV photons with a sun-like wavelength profile, under a large range of temperatures. Inactivation decreases with decreasing temperature (Weber & Greenberg, 1985; Dose & Klein, 1996; Noell et al., 2015). Inactivation follows an exponential behavior with a slight leveling off at higher fluence, likely due to spore clumping, a complex inactivation/repair mechanism, or small resistant subpopulations. Fitting the kinetics allows us to extrapolate the inactivation rate at various fluence and temperature. We expect *B. subtilis* spores to be deactivated by UV photons in a couple of hours and in a couple of days at the surface of Europa and Enceladus respectively, taking into account their distance to the Sun and average surface temperature.

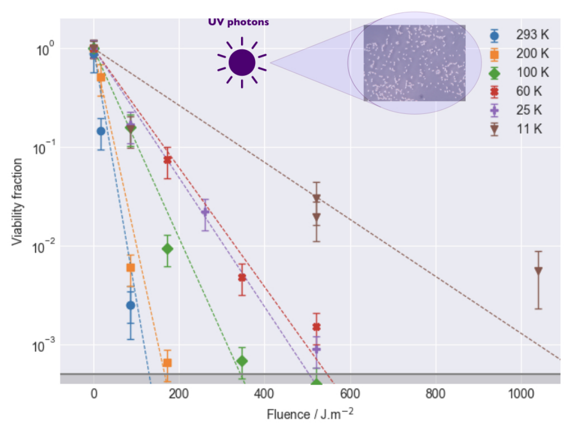


Figure 1: Viability fraction of *B. subtilis* spores under UV irradiation versus UV fluence for various temperatures and a phase contrast microscopy image of a sub-monolayer of *B. subtilis* spores deposited onto a glass slide, using a similar technique when performing UV-irradiations.

## 4. Future directions

Future investigations will focus on quantifying the effect of ice chemistry versus molecular shielding, and the impact of electron irradiation onto spore viability; while ice layers thicker than a couple of microns can shield bacterial spores from harmful UV radiations, energetic particles can create secondary low energy electron that generate radicals in the ice and potentially attack the spore coat. Direct interaction of electrons with the outer coat may also affect viability. In parallel, an DNA analysis and germination stage study of the spores will shed more light on the damage mechanism after irradiation at cryogenic temperatures.

## References

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