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# Subsurface Halophiles: An Analogue for Potential Life on Mars.

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Recent discoveries have reopened the idea that, in the past, Mars had a period of wetness where conditions were similar to those on Earth [1]. If this was the case then it is feasible that these environments may have harboured life. The martian surface today is dry, cold and heavily bombarded by UV radiation, making it an environment unsuitable for any known terrestrial life [2]; the martian subsurface might be considerably more hospitable. Subsurface microbial communities would not have access to sunlight for photosynthesis to drive their food chains, so primary production would have to be driven by chemolithoautotrophic organisms.

Every successful Mars landing site has been found to have abundant surface salt [3], and halite has been detected in Martian meteorites [4]. While subsurface halite deposits have not yet been detected on Mars, areas on the surface consisting of unidentified chlorine deposits have been detected with evaporation as one of the main theories to explain their creation [5]. On Earth, chloride evaporites are home to halophiles, and they have been suggested as an analogue for potential Martian life. Halophiles are micro-organisms which display a high salt tolerance. Despite being found in evaporite deposits, they are normally studied in surface brines. Brines form when the rate of evaporation of water is greater than the rate that water enters an area [6], so eventually most brines evaporate and form salt crystals. Halophiles in a brine are able to alter the size and formation rate of fluid inclusions within these salt crystals so as to entomb themselves inside until the crystals can re-dissolve [7]. It is uncertain how long halophiles can spend entombed within crystals, but there are some who speculate that it could be up to millions of years, if not longer [8].

If life had arisen on Mars during an earlier, more hospitable, wet period, as it did on Earth, then when the planet cooled, organisms analogous to terrestrial halophiles might have had the greatest potential for survival [9]. As the atmosphere left the planet, any large pools of water would have gradually evaporated, with the remaining fluid becoming increasingly brine-like. The only organisms that could survive these high salt conditions would have to be halophilic. The possibility exists that, even if there are no free-living martian halophiles remaining now, as the brines crystallized, halophiles could have been entombed inside mineral grains. It is feasible that their biomarkers and/or DNA could be extracted by instruments onboard future Mars landers [10].

It is also interesting to consider that, even though known terrestrial halophiles could not survive the high UV on the Martian surface today, they are still some of Earth's most UV tolerant terrestrial organisms. This is because, on Earth, briny environments tend to be created by evaporation of water brought on by intense sunlight; therefore organisms living in them get a high dose of UV [11]. It seems reasonable to suggest that a martian halophilic organism would, prior to the loss of Mars' atmosphere, initially have evolved in environments similar to the ones in which they evolved on Earth and so might share similar adaptations. There is some evidence to suggest that there is still periodic liquid water on Mars in the form of brines

[3]. Any surface-dwelling organism remaining on the planet would need to have UV and salt tolerance and be able to enter periods of dormancy during the long dry spells. Since terrestrial halophiles have all three of these attributes, they are probably good candidates for use as analogues of martian life.

Boulby Salt and Potash mine in Yorkshire is 1.4 km underground and the second deepest mine in Europe. Despite this depth and the darkness, Norton et al., isolated halophiles from the halite deposits [12]. In this project we will be attempting to isolate and characterize halophiles from halite, as well as from other salt-rich sediments such as potash, sylvinite, anhydrite and polyhalite, in order to gain an understanding of potential life in the subsurface of Mars. Although the Boulby mine is used as a martian analogue environment [13], it does possess certain key differences from modern Mars, in particular its aerobic environment. Our long-term goals, once we have characterized the micro-organisms present, is to expose them to Mars conditions (past and present) to determine their survival potential. We will also investigate differences between their growth in the mine and in the simulated martian brine environments. To detect the presence of potential extinct or extant life on Mars, we will also focus on attempting to define biomarkers that may be left by halophile growth and degradation in these Mars-like conditions.

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