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Fluorescence spectroscopy for the detection of life in the Salten Skov Mars regolith analogue

Hoi S. Chan, Zita Martins*, Mark A. Sephton

Department of Earth Science and Engineering, South Kensington Campus, Imperial College London, London SW7 2AZ, UK

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ABSTRACT

We have performed fluorescence spectroscopic analyses of the amino acid content of the Mars soil analog Salten Skov and of an amino acid standard mixture with the same amino acid abundance and distribution as the soil. This is the first study that investigates the total amino acid abundance of a Mars soil analog with a spectrofluorometer and correlates it with the results obtained from other analytical techniques. We discuss the variations between the fluorescence spectra of the Salten Skov soil sample and the amino acid standard mixture, and the differences between the Salten Skov hydrolyzed and non-hydrolyzed sample fractions. We also discuss the advantages and limitations of fluorescence spectroscopy in the context of future life detection missions.

1. Introduction

Ever since the collection of data from various missions to Mars, it has been broadly recognized that liquid water was, and may still be, present on the Red Planet (e.g. Anderson and Tice, 1979; Carr and Malin, 2000; Baker, 2001; Britt, 2004; Squyres et al., 2004; Haskin et al., 2005). Although liquid water is a crucial condition for life as we know, there is still no unambiguous evidence of life on Mars. An *in situ* study of the Martian environment is an extremely expensive and extravagant task requiring enormous background manpower, let alone the time spent on investigating and developing the mission and its associated instruments. Therefore, it is imperative to test the instrument on appropriate Mars soil analogs so as to facilitate the development of pertinent instrumentation that performs optimally in the challenging environment that may be encountered on Mars. Mars analogs provide a valuable preview of the behavior of Mars soil samples when examined using the potential rover instrumentation. They also provide important guidance for protocols or interpretative methods for distinguishing between biotic and abiotic materials.

Amino acids are amongst the most popularly targeted biomarkers owing to their roles as key chemicals in biochemistry. In this study the amino acid content of a Danish Mars soil analog – Salten Skov – was analyzed for the first time using a fluorescence detection technique. Salten Skov has a high organic content (Garry et al., 2006, Peeters et al., 2009), which is a valuable attribute when attempting to demonstrate the detection of subtle variations in amino acid contents using different analytical techniques. The costly *in situ* exploration of Mars surface requires careful selection of analytical locations. Potential sampling sites including pockets of organic-rich zones where situations are favorable for material scavenging or organics accumulation, or the subsurface where microbial life may be protected from the ionizing radiation. In the context of organic-rich pockets, Salten Skov is an excellent analog that allows cross-boundary analyses for its magnetic, organic, physical, and chemical traits, which also makes possible the identification of relationships between its organic content and other physical properties. This study compares the Salten Skov sample with its amino acid standard mixture counterpart using fluorescence detection, and aims to identify the variations between the amino acid content of the Martian analog before and after hydrolysis treatment (i.e. its free and total amino acid content, respectively). The amino acid standard mixture was made in accordance to the High Performance Liquid Chromatography-fluorescence detection (HPLC-FD) data retrieved in a previous study (Garry et al., 2006). This research, therefore, provides a means of comparing the effectiveness of state-of-the-art life detection methods.

1.1. Regional geology of the sampling site: mid-Jutland, Denmark

Soil samples collected in Salten Skov (Jutland, Denmark) have been recognized as one of the various analogs that can resemble

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[*] Corresponding author. Tel.: +44 20 7594 6473.
E-mail address: z.martins@imperial.ac.uk (Z. Martins).

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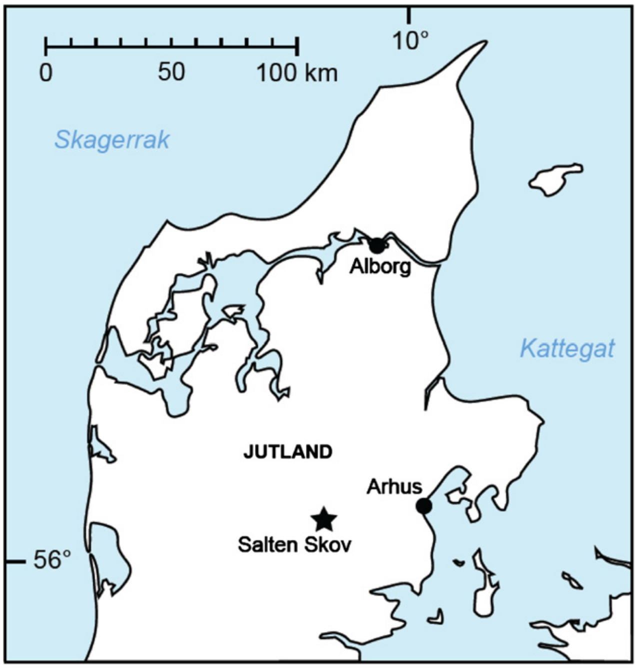

Fig. 1. Map showing the location of the Salten Skov sample site.

the Martian soil (e.g. Gunnlaugsson et al., 2002; Merrison et al., 2004; Nørnberg et al., 2004; Garry et al., 2006; Nørnberg et al., 2008; Marlow et al., 2008, 2011; Fig. 1). The Salten Skov deposit extends over several hundred square meters in Mid-Jutland, where 20 cm of reddish soil (Munsell color system: 2.5 YR 3/4) overlays 45 cm of a more yellowish (5 YR 4/6) sediment (Nørnberg et al., 2004). Several studies have investigated their mineralogical and magnetic properties (e.g. Gunnlaugsson et al., 2002; Merrison et al., 2004; Nørnberg et al., 2004), but thorough chemical studies are restricted to just a couple of papers (e.g. Garry et al., 2006; Peeters et al., 2009). The current study analyses for the first time the amino acid content of the Salten Skov soil using a spectrofluorometer.

The magnetism is contributed by the magnetic minerals in the soil, such as hematite, maghemite, and antiferromagnetic goethite (Nørnberg et al. 2004). A few papers have attempted to account for the origin of these ferrous minerals. Burning of soil, which is believed to be one of the mechanisms, dehydrates goethite into hematite and maghemite in Næsset, Denmark. Also, the red colouration (around 5 YR, 2.5 YR) usually indicates the presence of hematite in soil, possibly generated by the heating of goethite (Nørnberg et al., 2004). However, as there is no historic or prehistoric evidence of heating in this area, there are several other hypotheses attempting to explain the magnetic properties of Salten Skov, including organic processes (Gunnlaugsson et al., 2002) and precipitation of ferrihydrite from Fe^{2+} bearing groundwater (Nørnberg et al., 2008).

1.2. Physical and chemical properties of Salten Skov sediment

The Salten Skov sediment is an interesting Mars soil analog. Amongst its various properties, the fine particle size and magnetic traits are the ones that best mimic the Martian dust.

The Salten Skov soil has a low bulk density ($\sim$1.1 g/cm^3) that is similar to the Martian dust (Nørnberg et al., 2004). Analytical measurements reveal that the density of the drifting Martian material close to the landing site of the Viking Lander 1 was about 1.2 g/cm^3 (Moore and Jakosky, 1989). Salten Skov has a general particle size of around 63 μm, which can further be disaggregated into individual grains of around 1–2 μm when soaked in an ultrasound bath (Gunnlaugsson et al., 2002; Garry et al., 2006). In general the dust grains tend to cohere into aggregates of diameter greater than 1 mm due to their electrical charge of around 10^5e (Merrison et al., 2004). The top soil used in this study comprises mainly fine grained material (grain size smaller than 2 mm). 35.5% of this fine grained portion is composed of silt and clay size material (25.4% of silt size fraction (63–2 μm) and 10.1% of clay size fraction ($<$2 μm); Nørnberg et al. 2004). Martian soil, on the other hand, is also composed of clay size material. According to Nørnberg et al. (2008), the grain size of the Martian top soil has a mean diameter of 3 μm. The 10.1 wt% clay composition in Salten Skov represents a significant surface area considering the small sizes of the clay particles. This is a vital attribute to the stability of organic molecules such as amino acids since regolith particles can serve as physical shields to lethal UV radiations on Mars surface. As discussed also by Peeters et al. (2009), amino acids were found relatively more stable in Salten Skov under simulated Martian conditions when compared to other Martian soil analogs, as clays are able to prevent organics from being degraded. The fine grain size of the Salten Skov particles is of utmost importance to aid in our understanding of possible organic-hosting regions under Martian conditions.

Another crucial property of Salten Skov as a Mars soil analog is its magnetism, which is contributed by the ferrous magnetic minerals that comprise most of the soil (Nørnberg et al., 2004; 2008). An understanding of the magnetic properties of the analog can enhance functional safety when operating instruments on the surface of Mars. The mineralogy of the soil has been studied before, and was indicated to compose chiefly of goethite (α-FeO(OH); $\sim$75%), hematite (α-Fe_2O_3; $\sim$19%) and maghemite (γ-Fe_2O_3; $\sim$6%; Nørnberg et al., 2008). The low saturation magnetization of hematite ($\sigma_S \sim$0.4 Am^2/kg) is counter-balanced by the strong magnetic component from maghemite ($\sigma_S \sim$70 Am^2/kg), even though maghemite represented a minor portion of the soil (Nørnberg et al., 2004). The high concentration of these iron oxides reflects the high saturation magnetization of 3.9 Am^2/kg, albeit factors including the variation in particle aggregation and mass density might affect the estimations. Despite the fact that the saturation magnetization of Salten Skov is a little higher than that of the Martian regolith ($\sim$2 Am^2/kg), the soil still possess the magnetic properties that make it stand out from other Martian analogs. Therefore, Salten Skov is an ideal Martian analog in regards to the interesting relationships between its physical, magnetic, and organic properties.

1.3. Organic content of Salten Skov

The surface of Mars is relatively depleted in organic content, as illustrated by the lack of organic compounds detected by the Viking spacecraft (Biemann et al., 1976). Several analogs (e.g. desert soils from Arequipa region in Southern Peru and the Atacama Desert in Northern Chile) are also almost devoid of organic matter and are optimal organic analogs for testing life-detection instrumentations that aim to detect trace level of organic compounds on Mars. For these analogs, the amino acids abundances are primarily in the parts per billion (ppb) range (Navarro-González et al., 2003; Peeters et al., 2009), which demonstrates a low organic content on a par with the sterile Martian soil (Biemann et al., 1976).

Compared to the Martian soil, Salten Skov is significantly enriched in organic compounds and has high amino acid abundances (Garry et al., 2006; Peeters et al., 2009) and bacterial concentrations. The Salten Skov amino acid content is in the order of parts per million (ppm; Garry et al., 2006, Peeters et al., 2009). Although the high amino acid content was anticipated to be