POTENTIAL FOR TRIMETHYLSULFONIUM HYDROXIDE AS A THERMOCHEMOLYSIS REAGENT TO DETECT ORGANIC BIOMARKERS IN FUTURE LIFE-DETECTION MISSIONS. L. Kivrak[1], S. Halbert[1], A.J. Williams[1], A. Buch[2], Y. He[2], S.S. Johnson[3], K. Benison[4], S. Shaner[5], L.E. Judge[5], [1]Department of Geological Sciences, University of Florida, 241 Williamson Hall, PO Box , 112120 Gainesville, FL 32611 (lkivrak@ufl.edu)., [2]Department of Chemical Engineering, CentraleSupalec. [3]Department of Biology, Georgetown University [4]West Virginia University, [5]University of New Mexico

Introduction: The search for life on Mars centers on the detection and analysis of organic molecules. Organic molecules are the building blocks of all known life on Earth, and specific classes and distribution patterns of organic molecules preserved in the rock record on Earth can indicate the presence of past or present life [1]. For example, fatty acids are ubiquitous and abundant in microbial membranes, and can be preserved in the rock record for up to several million years [2]. Life on Earth predominantly synthesizes even-chained fatty acids, leading to a predominance of even over odd chained fatty acids that could provide a reliable signal distinct from the background of abiotically produced organics [3].

Landers and rovers on Mars from Viking to Perseverance have carried instruments designed to detect organic molecules [4] such as gas chromatography-mass spectrometry (GC-MS) sometimes combined with pyrolysis (Py-GC-MS). GC-MS volatilizes and separates organic molecules in a time series, then measures the mass of component ions using a mass spectrometer, allowing organic molecules to be identified with a high degree of specificity [4].

The Curiosity rover contains the most advanced GC-MS yet used on another planet: the Sample Analysis at Mars (SAM) instrument [4]. SAM has the capability to perform Py-GC-MS, as well as wet chemistry experiments. A wet chemistry experiment such as thermochemolysis or derivatization can be used to improve the yield of organic matter by transforming polar molecules into nonpolar volatile derivatives that are more easily detectable by the GC-MS [5].

During thermochemolysis, the reagent tetramethylammonium hydroxide (TMAH) ruptures hydrolysable bonds and deprotonates acidic groups [6]. In fatty acids, this leads to the formation of fatty acid methyl esters (FAMES). TMAH is present on the SAM instrument and has been used to detect organic compounds in Gale Crater, including benzothiophene and benzoic acid [7]. Trimethylsulfonium hydroxide (TMSH) thermochemolysis works via a similar chemical mechanism to TMAH [8]. It has been used on Earth for the analysis of phenols, pharmaceutical drugs, herbicides, and phenolic wood composites [9] but has never flown on a spacecraft. The C-S bonds in TMSH are weaker than the C-N bonds in TMAH, causing the decomposition of TMSH at lower temperatures, which leads to methylation of organic compounds [10]

Here we examine the utility of TMSH as a potential thermochemolysis reagent for use in future GC-MS bearing life detection missions. We examine the optimal pyrolysis temperature for the use of TMSH thermochemolysis on organic chemical standards of nucleobases and fatty acids, as well as on planetary analog samples.

Samples: *Organic Standards:* Nucleobase standards of adenine, thymine, cytosine, guanine, and uracil, as well as the metabolic intermediaries hypoxanthine and xanthine were made at 0.25 mol/L in TMSH (25% in methanol). A mixture was made at 0.25 mol/L of each nucleobase. A Fatty acid mixture of C_{14}, C_{16}, C_{18}, C_{20}, C_{22}, and $C_{22:1}$, at 1g/L each was pyrolyzed at 350, 400, 500, and 600°C

Analog Samples: The sample suite here is analogous to environments on Mars such as the hydrothermal sinter detections in Gusev Crater [11], and clay and sulfate-rich lacustrine sediments of Gale crater [12]. The samples include actively and recently formed siliceous sinter from a hydrothermal system in Iceland [13], modern schwertmannite precipitates from Iron Mountain, California, and core samples from ephemeral sulfur-acidic lakes.

Methods: *Organic Standards:* 3 µL of sample was added to solvent washed sample cup, along with 3 µL of TMSH and 1.5 µL naphthalene-d8 standard for quantification. Sample was pyrolyzed at 350, 400, 500, 600ºC. The GC-MS program ramped from 50ºC (5 minute hold) 50-240ºC with a 6ºC/min ramp, 240-300ºC with a 10ºC/min ramp after the methods of [14].

Analog Samples: 3-5 mg of powdered sample was placed into sample cups with 1.5 µL of C_{19} as an internal standard. TMSH or TMAH was added to each samples cup at a ratio of 1µL TMSH/TMAH to 1mg sample. Samples were pyrolyzed at 600ºC. Several of the analog samples were also pyrolyzed at 350, 400, and 500ºC to better understand the effect of pyrolysis temperature. The oven program ramped from 50°C to 300°C at 20°C/min with a 10 minute hold.

Results and Interpretations:

Organic Chemical Standards: The yield of detected nucleobases from TMSH thermochemolysis increased with increased pyrolysis temperature. Significant

quantities of identifiable nucleobases were present at all temperatures. In a mixture of nucleobases, all nucleobases identified with TMAH thermochemolysis by [14] were detected using TMSH thermochemolysis. Several nucleobases derivatives that were not detected with TMAH thermochemolysis, such as dimethyl cytosine, dimethyl adenine, and guanine derivatives were detected with TMSH thermochemolysis in the mixture. At lower temperatures, TMSH was a distinct improvement over TMAH results from [14]. In the fatty acid mixture, a pyrolysis temperature of 400ºC provided the highest yield. These results consistently show that the ideal pyrolysis temperature for TMSH thermochemolysis is 400-500ºC, lower than that of TMAH thermochemolysis. This trend was also seen in the analog samples. A silica sinter hydrothermal sample (I9) and an Iron Mountain Schwertmannite sample (SS12) were pyrolyzed at multiple temperatures with TMSH thermochemolysis. In both samples, the quantity of detected FAMES was lowest at a pyrolysis temperature of 600ºC, and highest at 500 or 400ºC.

Iceland Sinter: The sinter sample from the recently active hydrothermal vent (I6) was pyrolyzed at 600ºC, and detections of C_{14}, C_{16}, C_{17}, C_{18}, and C_{20} were made with TMSH thermochemolysis. TMSH and TMAH thermochemolysis of the active hydrothermal sample (I9) resulted in the detection of C_9, C_{10}, C_{12}, C_{14}, C_{16}, C_{18}, and C_{20}.

Iron Mountain Schwertmannite: TMSH thermochemolysis at 600ºC in schwertmannite samples SS6 and SS10 did not result in any FAME detections. However, TMSH thermochemolysis of SS2 at 600ºC resulted in the detection of C_8, C_9, C_{10}, C_{12}, C_{14}, C_{16}, C_{18}, and C_{20} FAMES. It is likely that the high iron-sulfide content and the acidity of these samples reduced the effectiveness of TMSH thermochemolysis. However, given that the yield of detected FAMES in SS12 improved with decreased pyrolysis temperature, future analyses will involve the pyrolysis of SS6 and SS10 at lower temperatures with TMSH thermochemolysis.

Australia Lacustrine Sulfur-Rich Samples: TMSH thermochemolysis at a pyrolysis temperature of 600ºC of SSJ2, a modern highly acidic sample, resulted in the detections of C_9, C_{12}, C_{14}, C_{16}, and C_{18}. TMAH thermochemolysis at 600ºC resulted in the detection of a multitude of alkenes and branched moieties, but no FAMES. TMSH thermochemolysis of SSJ5 at 600ºC , a modern acidic sample, with clay minerals and goethite, resulted in the detection of C_{12}, C_{14}, C_{16}, and C_{18}. TMAH thermochemolysis of SSJ5 at 600ºC resulted in the detection of the same FAMEs, with potential identifications of C_8, C_9, and C_{10}. SSJ3 and SSJ4 were circumneutral clay-rich samples. TMSH thermochemolysis of SSJ3 resulted in the detection of C_{14}, C_{16}, and C_{18} FAMES, while TMAH thermochemolysis resulted in the detection of the same FAMES, in addition to several alkanes. TMSH thermochemolysis of SSJ4 resulted in no FAME detections at a pyrolysis temperature of 600ºC. However, when the pyrolysis temperature was lowered to 400ºC, the detection of a multitude of FAMES from C_{10}-C_{30} was possible.

Table 1: FAME detections with TMSH thermochemolysis at 600 ºC ('o'= detection)

		C8	C9	C10	C12	C14	C16	C17	C18	C20
Sinter	IC09		o	o	o	o	o		o	o
	ICS6					o	o	o	o	o
Fe-oxides	SS12	o	o		o	o	o		o	
	SS10	none								
	SS8	none								
	SS6	none								
	SS2	o	o	o	o	o	o		o	o
Australia Lake Sediments	SSJ2		o		o	o	o		o	
	SSJ3					o	o		o	
	SSJ4	none								
	SSJ5				o	o	o		o	

Conclusion: TMSH is a thermochemolysis reagent with great potential for use in future life-detection missions. We have shown its utility for FAME detection in Mars analog samples. In many of the samples here, TMSH thermochemolysis resulted in the same FAME detections as TMAH thermochemolysis. Additionally, we have demonstrated that pyrolysis temperature is a key factor in the use of TMSH thermochemolysis. Acidic iron and sulfur bearing samples react poorly with TMSH thermochemolysis, especially at higher pyrolysis temperatures. Future work will further explore the relationship between sample acidity, mineralogy, and TMSH thermochemolysis, and will include analyses of analog samples of other planetary bodies.

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