Solid Phase Micro Extraction: Potential for Organic Contamination Control for Planetary Protection of Life Detection Missions to the Icy Moons of the Outer Solar System

Samuel H. Royle[1]; Jonathan S Watson; Yuting Zhang; Georgios Chatzitheoklitos; Mark A. Sephton

Impacts and Astromaterials Research Centre, Earth Science and Engineering, South Kensington Campus, Imperial College London, SW7 2BP

Corresponding author: Samuel H. Royle; Impacts and Astromaterials Research Centre, Earth Science and Engineering, South Kensington Campus, Imperial College London, SW7 2BP; +44 (0)20 7594 9981

s.royle@imperial.ac.uk

Running title: SPME: Planetary Protection of Icy Moons

1. Abstract

Conclusively detecting, or ruling out the possibility of, life on the icy moons of the outer solar system will require spacecraft missions to undergo rigorous planetary protection and contamination control procedures to achieve

extremely low levels of organic terrestrial contamination. Contamination control is necessary to avoid forward contamination of the body of interest and to avoid the detection of false positive signals which could either mask indigenous organic chemistry of interest or cause an astrobiological false alarm. Here we test a new method for rapidly and inexpensively assessing the organic cleanliness of spaceflight hardware surfaces using solid phase micro extraction (SPME) fibres to directly swab surfaces. The results suggest that the method is both time and cost efficient. The SPME-gas chromatography mass spectrometry (GC-MS) method is sensitive to common mid-weight, non-polar contaminant compounds, e.g. aliphatic and aromatic hydrocarbons, which are common contaminants in laboratory settings. While we demonstrate the potential of SPME for surface sampling, the GC-MS instrumentation restricts the SPME-GC-MS technique's sensitivity to larger polar and non-volatile compounds. Although not used in this study, to increase the potential range of detectable compounds, SPME can also be used in conjunction with high performance liquid chromatography/liquid chromatography-mass spectrometry systems suitable for polar analytes [*Kataoka et al.*, 2000]. Thus, our SPME method presents an opportunity to monitor organic contamination in a relatively rapid and routine way that produces information-rich data sets.

Key Words: SPME; Planetary Protection; Organic Contamination; Life Detection; Icy Moons

2. Introduction

Numerous past, current and future space missions had and have the detection of extraterrestrial organic matter as a primary goal. Conclusive detection requires the avoidance of forward contamination of the bodies of interest. An extremely low-level of organic (both biological and non-biological) terrestrial contamination is necessary; which translates to a high level of cleanliness for all parts of the spacecraft that may come into contact with samples for analysis [e.g. *Blakkolb et al.,* 2014].

Minimising the contributions of organic contaminants, termed 'contamination control', is required for life detection missions. Recently the search for life has turned towards the icy moons of the Jovian and Saturnian systems [*Reynolds et al.,* 1983; *Task Group on the Forward Contamination of Europa*, 2000; *McKay et al.,* 2008; *Parkinson et al.*, 2008]. False positives (indicating life or pre-biotic chemistry) caused by the detection of organic contaminants could either mask indigenous organic chemistry of interest or cause an astrobiological false alarm leading to unwarranted stringent planetary protection requirements for future missions [*Task Group on the Forward Contamination of Europa*, 2000; *Mahaffy et al.,* 2003]. It should also be noted that avoiding forward contamination is also important for the reliability of analytical results from organic matter detection missions exploring bodies where we do not expect to find evidence of past or present life, such as those exploring asteroid or cometary bodies [*Drake et al.,* 2011; *Nakamura et al.,* 2012; *Tsuda et al.,* 2013; *Westphal et al.,* 2014; *Lauretta et al.,* 2015].

Contamination knowledge includes the documentation of all known potential organic contaminants to ensure contaminating molecules are not mistaken for compounds of interest during sample analysis. It is useful to define contaminants, in a planetary protection contamination control sense, as those substances that can be detected and that could cause issues for the current life detection mission. As such, contamination control for astrobiology missions may focus on a limited range of key compounds (Table 1).

Organic contamination control in respect to planetary protection is only concerned with affecting the results of the current science mission and is not concerned about contaminating the body itself and the effects on future science missions because, unlike biological contamination control, the compounds of interest are not expected to be self-replicable and would, most probably, stay localised or if dispersed in a liquid medium, such as the subsurface ocean of an icy moon, would simply dilute to undetectable concentrations.

Spacecraft are typically cleaned to a non-volatile residue cleanliness of 1 $\mu g/cm^2$ which, based on the IEST-STD-CC1246D standard, is level A. For astrobiologically-sensitive parts of a mission, such as the Mars Science Laboratory (MSL) sample handling chain [*Mahaffy et al.*, 2003], 100 ng/cm^2 or level A10 is the standard. The Viking sample handling hardware was cleaned to 1 ng/cm^2 or Level A1000 – although this is an extreme example, it may well be the level necessary for life detection missions where the types and

concentrations of analytes are uncertain, such as with missions to the icy moons.

To achieve these high levels of cleanliness a variety of techniques are employed:

All work is carried out under clean room conditions. A typical aerospace cleanroom is class 100,000 or ISO 8 (i.e. contains 100,000 or fewer particles of 0.5 μm in diameter per cubic foot of air, controlled by High Efficiency Particle Air (HEPA) filters and maintaining a positive air pressure between the inside and the outside environments. For the assembly of the most sensitive flight hardware, class 10 or ISO 4 cleanrooms may be employed, containing less than 10 0.5 μm particles per cubic foot of air and in these environments personnel must be isolated by wearing clean suits at all times.

Precision cleaning is the series of processes targeted at removing both particles and molecular films of organic contaminants. Firstly, visible contamination is wiped from the surface. Secondly, a series of rinses with organic and aqueous solvents of varying polarities is performed – which may be coupled with ultrasonic treatment to liberate any contaminants adhering to the surfaces. Thirdly, Freon vapour is used for degreasing. Fourthly, isopropyl alcohol rinses are performed and analysed for remaining particulate levels. The overall process of precision cleaning commonly achieves Level 100 cleanliness, high enough in most cases [*Mahaffy et al.*, 2003]. The use of solvents is not suitable for all materials used in spacecraft assembly (e.g. plastics and polymers can be

dissolved by some solvents) so other techniques are also employed in these cases, using plasma, accelerated CO_2 snow, radiation or electron beams to remove organic compounds [e.g. *Task Group on the Forward Contamination of Europa*, 2000; *Committee on preventing the forward contamination of Mars*, 2006; *ten Kate et al.*, 2008; *Dworkin et al.*, 2017]. Repeated wiping of surfaces with clean room cloths saturated with isopropyl or ethyl alcohol during assembly prevents the re-build-up of molecular contaminants.

Thermal bakeout is the concluding step to both remove surface contaminants and reduce subsequent outgassing of organic impurities within the materials. Depending on the material, bakeouts may range from 70 °C to 105 °C and last 72 to over 160 hours. The Viking One Lander was baked out after the final assembly as a terminal sterilization step at 112 °C for 30 hours [*Martin*, 1975].

Monitoring clean room and hardware organic cleanliness is currently a complex process often involving multiple, expensive and time-consuming techniques. [*Task Group on the Forward Contamination of Europa*, 2000; *Mahaffy et al.*, 2003; *Blakkolb et al.*, 2014; *Li et al.*, 2015]. Effective monitoring is confounded further by the fact that, in contrast to biological contamination, there are currently no strictly defined quantitative limits for organic contamination control. The lack of well-defined guidance is partly due to the fact that the cleanliness level has to be appropriate to the sensitivity of the instruments of the specific mission. If the instrument cannot detect the contaminants they are not an issue. However, initially undetectable

compounds may be transformed into more problematic species by the harsh environments encountered during the long cruise phase, in-orbit or (if applicable), once landed on the surface. This transformation of organic molecules into other, more problematic, species is particularly a problem for the moons of the Jovian and Saturnian systems due to the increased and highly variable radiation environment the spacecraft will encounter throughout the life of the mission and the highly oxidative surface environment if the mission incorporates a landed element [e.g. *Cassidy et al.*, 2010; *Johnson et al.*, 2012; *Kimura and Kitadai*, 2015]. With the numerous unknowns associated with exploration of the icy moons, this potential complication highlights the importance of cleanliness.

The OSIRIS-REx sample return mission to the asteroid Bennu had a strict contamination control plan. Procedures to limit the total contamination burden on the returned sample were put in place to limit sensitive surfaces to cleanliness levels (established in IEST-STD-CC1246D) at the 100A/2 level [*Borson*, 2005]. In addition, due to the unique mission science objectives, specific contaminants of concern were limited to a total accumulation of 180 ng/cm^2 on the most sensitive surfaces of the sample handling chain [*Lauretta et al.*, 2017]. While not planetary protection-related, optical instruments often have the most stringent organic contamination limits. For instance, for the Hubble Wide Field Planetary Camera, recontamination is limited to a rate of

47 ng/cm^2/month and the camera undergoes a monthly decontamination cycle that reduces the level to 1 ng/cm^2 [*Hedgeland et al.,* 1994].

Good examples of organic cleanliness monitoring are available from missions to Mars. Multi-stepped solvent extraction, followed by pre-concentration of analytes (by evaporation) and analysis by diffuse reflectance infrared Fourier transform (DRIFT) spectroscopy, Fourier transform infrared (FTIR) spectroscopy and pyrolysis-gas chromatography-mass spectrometry (Py-GC-MS) techniques were carried out on swabs from surfaces of the MSL sample transfer chain hardware at various stage of construction [*Blakkolb et al.*, 2014]. The use of multiple solvents, however, complicated the analysis of the data and diluted the contaminants of interest, reducing sensitivity of the detection. The whole process of extraction, concentration and analysis was also very time consuming and therefore costly. A similar process to that employed for MSL is proposed for Mars 2020 [Table 2, *Summons et al.*, 2014]

Various culture dependant assays [*Benardini Iii et al.*, 2014a, 2014b] and culture independent methods (such as 16s RNA-based diversity; next generation sequencing; viability-linked metagenomics assays (propidium monoazide treatment; quantitative polymerase chain reaction) [*La Duc et al.*, 2004, 2009; *Nellen et al.*, 2006; *Probst et al.*, 2012] have been used to track the microbial bioburden present on flight instrument surfaces. While the NASA/ESA standard assay technique [*Morris et al.*, 2010] is a good example of a standardised planetary protection contamination control method that is

missing from non-biological contamination control, these techniques give no indication of the non-biological organic contamination present.

A diagnostic organic contamination monitoring process is needed. While useful, witness plates [*ten Kate et al.*, 2008] can only show what is condensing/falling onto clean metal surfaces, they cannot show transfer from hands/gloves as they are not handled in the same way as the actual flight hardware. A standardised technique to directly sample the flight hardware surfaces in addition to the atmosphere itself, which is rapid, inexpensive and easy to use would be very useful (alongside the use of witness materials) in keeping track of clean room cleanliness on a regular basis.

Solid phase micro extraction (SPME) is a sample preparation method developed for the analysis of organic compounds [*Arthur and Pawliszyn*, 1990]. In the analysis for organic compounds by SPME, a fused silica optical fibre coated with liquid organic polymer or solid sorbent is exposed to the sample matrix wherein a distribution equilibrium of the analytes is established between the matrix and the coating; this combines sampling and pre-concentration of analytes into a single step [*Harper*, 2000]. The analytes collected are thermally desorbed, in the injector of a gas chromatograph for analysis. SPME is advantageous for the analysis of organic compounds due to its high speed (extraction time can be reduced to a few minutes instead of the hours/days of classical liquid-liquid extraction methods), low cost, elimination of solvents from both the extraction and analysis steps (although a solvent may

195 be used instead of thermal desorption for analysis [*Arthur and Pawliszyn*,
196 1990]), portability, applicability to gaseous, liquid or solid samples and
197 relative independence of destined analytical instrument design [*Arthur and*
198 *Pawliszyn*, 1990; *Louch et al.*, 1992; *Otu and Pawliszyn*, 1993].

199 In this study we assessed whether SPME could be employed as a standardised
200 inexpensive, rapid and accurate technique for monitoring the lab atmosphere
201 and flight hardware surfaces for organic contamination control and general
202 cleanliness for future life and organic matter detection space missions. We
203 developed a method to effectively sample hardware surfaces. We assessed
204 sensitivity for detecting organic compounds that have been identified as
205 contaminants of interest for astrobiological missions. This study is timely as
206 there are currently numerous life and/or organic matter detection missions to
207 the icy moons of the outer solar system in various stages of planning and
208 implementation [Powell et al., 2005; Erd, 2012; Pappalardo et al., 2013;
209 Dachwald et al., 2014; Phillips and Pappalardo, 2014; Konstantinidis et al.,
210 2015].

211 **3. Materials and Methods**

212 *3.1. SPME fibre selection and sampling procedure development*

213 We chose 30 µm coating thickness polydimethylsiloxane (PDMS) coated
214 SPME fibres (Supelco, USA) as these were suited to detecting the greatest
215 range of non-polar compounds of interest. Other SPME fibre types with
216 different coatings and coating thicknesses could be selected for more specific

217 contaminants of interest based on the manufacturer's recommended usage
218 (Table 3).

219 *3.2. SPME fibre preparation*

220 SPME fibres were held in a manual SPME holder. Prior to the use of the
221 SPME fibres they were conditioned by heating in the inlet of the GC for 45
222 minutes at 300 °C.

223 *3.3. Standard compound selection*

224 Standard organic compounds of interest were selected based on those
225 identified as problematic for astrobiological missions by Mahaffy et al. (2003),
226 Table 1. Only the non-polar compounds were selected to be tested as the more
227 polar compounds would require techniques such as SPME-high performance
228 liquid chromatography (SPME-HPLC) or SPME-liquid chromatography-mass
229 spectrometry (SPME-LC-MS) [*Kataoka et al.*, 2000], which is beyond the
230 scope of this study.

231 The mid-length C_{18} alkene and alkane compounds 1-octadecene and
232 octadecane, three ring polycyclic aromatic hydrocarbon (PAH) phenanthrene
233 ($C_{14}H_{10}$), the saturated C_{18} fatty acid octadecanoic acid ($CH_3(CH_2)_{16}COOH$),
234 the triterpene squalene ($C_{30}H_{50}$) and the sterol cholesterol ($C_{27}H_{46}O$) (Figure 1)
235 were selected for their relatively low volatility and non-polarity (to varying
236 degrees).

237 *3.4. Surface spiking*

The test surfaces used were the flat portions of a 316 stainless steel cap from Swagelok® (part number SS-20M0-C).

Stainless steel surfaces were prepared for spiking by sonicating the Swagelok® caps in propan-2-ol (isopropyl alcohol; IPA) (HPLC plus grade 99.9%, Sigma-Aldrich) for 10 minutes and then heating them overnight (minimum 15 hours) at 125 °C, to replicate dry heat microbial reduction (DHMR) and 'bake out' any initial organic contaminants [*Pflug*, 1971].

After cleaning, the stainless steel samples were only handled with nitrile gloves that had been wiped with IPA, with no contact being made with the surfaces for analysis. The samples were also kept wrapped in aluminium foil (which had undergone the same overnight heat treatment) between experimental steps to prevent fallout of airborne contaminants onto the surfaces.

10 µl of the solutions of the individual standard organic compounds in dichloromethane (DCM; 99.8+ % distol-pesticide reagent grade, Fisher Scientific) were syringed onto the stainless steel surface to give contamination levels corresponding to A (1000 ng/cm^2), A10 (100 ng/cm^2), A100 (10 ng/cm^2), A1000 (1 ng/cm^2) and the DCM was allowed to fully evaporate.

During the external standard calibration phase of the experimental procedure, if a compound was below the limit of detection (<LOD) at an analysed concentration by liquid injection, the relevant contamination level was not tested (nor contamination levels below that) for SPME-GC-MS sensitivity, as

sensitivity to that compound at astrobiologically relevant levels was shown to be limited by the GC-MS method.

3.5. SPME sampling procedure

Prior to swabbing 5 µl of IPA was syringed onto the test surface. The activated SPME fibre was used to rub over this 'wetted' test surface to aid transfer of IPA soluble contaminants from the surface to the fibre

Each experiment was carried out 4 times to test reproducibility of the technique.

Experimental blanks (referred to in the results as 'surface blanks') were performed by carrying out the sampling procedure on the test surface after the cleaning procedure, without spiking with the standard compounds.

3.6. SPME-Gas Chromatography-Mass Spectrometry (SPME-GC-MS)

Analysis of the volatiles adsorbed onto the SPME fibre was carried out via SPME-GC-MS analysis. The SPME fibre was inserted directly into the injector of a Perkin Elmer Clarus 580 gas chromatograph coupled to a Clarus SQ85 mass spectrometer (GC-MS). Analytes were desorbed from the SPME fibre for 10 minutes into the injector, which was operated in split mode with a 20:1 split ratio and held at 290 °C, with a column flow rate of 1.1 ml min^{-1}. Separation was performed on a J&W DB-5 ((5%-Phenyl)-methylpolysiloxane) column (30 m x 250 µm x 0.25 µn). The GC oven was held for 2 min at 60 °C and then ramped at a rate of 10 °C min^{-1} to 310 °C where it was held for 5 min.

Mass spectra were acquired simultaneously in full scan (45-550 m/z) and selective ion monitoring (SIM), the ions detected were m/z 55, 57, 69, 73, 178, 217 with a dwell time of 50 ms. Recoveries were calculated from external standards injected in solution under the same conditions.

Analytical blanks were performed by the insertion of the SPME fibre into the injector of the GC-MS directly after activation.

4. Results

Integrated peak area in the extracted ion chromatogram of the characteristic ion fragment for that compound (m/z 55 = 1-octadecene; m/z 57 = octadecane and octadecanoic acid; m/z 69 = squalene; m/z 178 = phenanthrene; m/z 217 = cholesterol) was used as a proxy for relative detectability (data used for calculation shown in Table 4). Total ion chromatograms were used to confirm peak identity.

The SPME recovery was thereby calculated by comparing the peak areas produced via SPME-GC-MS (using the average of 4 SPME swabbings) against that which was expected based on the liquid injection calibration curve and expressing this as a percentage (Table 4).

SPME-GC-MS selected ion current chromatograms produced for all compounds are shown in Figures 2-7.

5. Discussion

5.1. Relative sensitivity of SPME-GC-MS relative to liquid injection GC-MS calibration

While recovery was reduced, in the majority of cases, if a standard organic compound was detected by the liquid injection technique, it was also detected by SPME-GC-MS at the equivalent surface spiking concentration. The only cases where SPME-GC-MS failed to recover a compound over the LOD which had been detected by liquid injection (at the relevant concentration) were at the lowest concentrations detectable (by liquid injection) for octadecane, 1-octadecane, octadecanoic acid and cholesterol. There are in fact low responses at the correct retention times in the SPME-GC-MS selected ion chromatograms for A1000 level octadecane and 1-octadecene and A1 level cholesterol that do not appear in the surface blanks. These low responses are, however, too weak to be quantifiable.

This all suggests that the limiting factor in this technique, at astrobiologically relevant contamination levels, is the sensitivity of the GC-MS technique used.

Variation in the percentage recovery by the SPME technique between compounds is likely due to the partition co-efficient between the compound, the IPA solvent and the fibre coating. The more polar compounds such as octadecanoic acid (with its polar carboxyl group) are less soluble in IPA and so more likely to stick to the stainless steel surface, reducing recovery.

5.2. Reproducibility – True positive rate (sensitivity)

Whilst quantitative reproducibility of the SPME-GC-MS technique was poor and standard deviation between repetitions was high (Table 4), qualitative reproducibility was good. A standard organic compound at a certain contamination level was either detected in all of the SPME swabs or none, this is expressed in terms of sensitivity.

Sensitivity and Specificity are statistical terms relevant in life detection missions in the solar system, and specifically those to the Icy Moons [*Sephton et al.*, 2018].

The sensitivity (or true positive rate) of a technique is its ability to make a correct detection of organic matter, this is investigated by calculating the rate of true positive detections in a sample with known composition.

$$Sensitivity = \frac{\#\ true\ positives}{\#\ true\ positives + \#false\ negatives}$$

Specificity (or the true negative rate) is the technique's ability to correctly identify a negative response, i.e. not detecting a false positive in a blank sample.

$$Specificity = \frac{\#\ true\ negatives}{\#\ true\ negatives + \#false\ positives}$$

A true negative represents a null detection in a sample that contains nothing, an example is the surface blanks after the steel test surface had undergone the cleaning procedures. A null detection in this context is a sample analysis producing no detectable compounds other than the IPA used in the method.

As all surface blanks showed no detectable compounds the specificity of the technique for surfaces that have undergone contamination control was 100 %.

Measurements of the spiked test surfaces were used to investigate the sensitivity of the technique for different standard compounds and concentrations. In this case, detecting the standard compound, e.g. squalene, on the spiked test surface would represent a true positive whereas not detecting the standard compound would represent a false negative (as it is known to be present). Sensitivity of the SPME sampling technique was found to be highly variable between the compounds tested and the results are shown in table 5.

However, as sensitivity was either 100 % or 0 % (compounds were either detected in all 4 replicates or not at all) the reproducibility of the SPME-GC-MS technique was excellent.

5.3. Standard organic compound relative selectivity and contamination control relevance

The SPME-GC-MS technique tested was found to have variable sensitivity to the different standard organic compounds tested. Based on the characteristics of the molecules (Table 6) it is possible to say something about the potential of this technique in detecting different classes of compounds relevant to organic contamination control for planetary protection.

The SPME-GC-MS method employed here proved very sensitive to phenanthrene from levels A to A1000 where there has been a 100 %

successful positive detection. Phenanthrene is a typical PAH, there is great interest in PAHs from an astrobiological point of view as evidence of their presence has been detected in meteorites [*Sephton*, 2002], atmospheric hazes [*Trainer et al.*, 2004] and even interstellar space [*Tielens*, 2008] so are important in understanding prebiotic chemistry. However, as PAHs are common products of combustion processes, especially diesel (and to a lesser extent) petrol exhaust emissions [*Haefliger et al.*, 2000; *Botta et al.*, 2008] they ae often recorded as contaminants in otherwise organically-lean environments/samples [*Botta et al.*, 2008; *Calaway et al.*, 2014]. PAHs are often a component of particulate contamination, falling out from the atmosphere onto surfaces [*Giger and Schaffner*, 1978; *Hodge et al.*, 2003], keeping track of particulate contamination on actual spaceflight surfaces throughout the build, as well as just on witness plates, is important to track change over time (effectiveness of cleaning procedures, etc.) and in case differences in molecular affinity to the surfaces or airflow patterns cause differential distribution of contaminants.

The SPME-GC-MS method also proved very sensitive to squalene from levels A to A1000 where there was a 100 % successful positive detection rate. This is likely to be as a result of squalene being reasonably volatile and non-polar. The effectiveness of the SPME-GC-MS technique in detecting squalene at a range of concentrations will be particularly useful in planetary protection contamination control. Squalene is one of the major components of human

sebum [*Kim and Karadeniz*, 2012] and a terrestrial contaminant attributed to human contamination. Human sebum will inevitably cause a false positive in life detection missions therefore effective monitoring to check the cleanliness of spacecraft hardware is fundamentally necessary. At a minimum, gloves that are wiped off with IPA should be worn at all times to avoid human contamination and this SPME-GC-MS technique proves a quick and effective way to check for accidental human recontamination of surfaces.

Octadecane and 1-octadecane are typical mid-chain length aliphatic hydrocarbons which are common contaminants of biological source, from the breakdown of biopolymers, for example from (terrestrial) microbial life [*Biller et al.*, 2015], organic oils such as those in lubricants [*Grosjean and Logan*, 2007], for example in vacuum pumps that are present in laboratory settings [*Illing et al.*, 2014], plastic polymers [*Grosjean and Logan*, 2007; *Brocks et al.*, 2008], diesel fumes [*Hauser and Pattison*, 2019]. The detection of pairs of alkenes and alkanes, especially at these longer chain lengths, could thus be taken as an indicator of biological activity if detected on mission leading to a false positive life detection. Hints of their presence in the A1000 level SPME-GC-MS selected ion chromatograms suggest that with a more sensitive GC-MS method (lower split ratio, etc.) these would be above detection limits even at the levels of contamination necessary in the most astrobiologically-sensitive areas of a spacecraft (A1000 level).

Octadecanoic acid, and other long chain length fatty acids are often used as biomarkers as they are indicative of a biological source [*O'leary*, 1962; *Volkman et al.*, 1989; *Alfaro et al.*, 2006; *Tan et al.*, 2018], however the polar carboxyl group makes detection via the GC-MS technique used difficult without derivatization of the molecule. Amino acids, a potential target biomarker of life-detection astrobiological missions and interesting in the context of icy moon prebiotic chemistry [*Elsila et al.*, 2009; *Martins and Sephton*, 2010; *Neish et al.*, 2010; *Johnson et al.*, 2012; *Dworkin et al.*, 2017] are more polar and so are not detectable by the column chromatography used, hence why they were not tested in the current study, however they too may be detectable by SPME-LC-MS or SPME-HPLC.

The lack of detection even at level A indicates that cholesterol does not desorb from the fibre, possibly due to its low volatility indicated by its large size and relatively high enthalpy of vaporization (Table 6). Large, low volatility compounds are less mobile and less likely to be transferred to surfaces in a clean lab environment and so may be not so important.

Thus the SPME-GC-MS technique employed is not suited to larger molecular weight compounds, like cholesterol, or those which are more polar, like fatty acids. However, if these larger-weight molecules are contaminants of concern for a particular mission, then SPME-LC-MS or SPME-HPLC need to be investigated to overcome the current limitations to small and volatile compounds due to the GC-MS instrumentation, potentially enabling the

detection of a much wider range of compounds at astrobiologically relevant concentrations.

Conclusion

- A new method that utilises SPME fibres to swab spacecraft hardware surfaces is demonstrated for the monitoring of cleanliness in planetary protection contaminant control procedures.
- The SPME-GC-MS method is convenient, both time and cost efficient. It can be employed into many stages of space missions.
- The SPME-GC-MS method is particularly sensitive to squalene and therefore human contamination at all levels tested.
- The SPME-GC-MS method is sensitive to common mid-weight, non-polar contaminant compounds e.g. aliphatic and aromatic hydrocarbons.
- The SPME-GC-MS method is not particularly sensitive to larger polar and non-volatile compounds as it is limited by the GC-MS instrumentation.
- While the potential of SPME for surface sampling is demonstrated here, future work needs to demonstrate the effectiveness of other desorbtion/detection techniques (SPME-LC-MS/HPLC) on specific compound classes, especially the higher molecular weight and polar species that are undetectable by the GC-MS technique employed here.

6. Acknowledgements

We would like to acknowledge the members of the Planetary Protection of the Outer Solar System (PPOSS) project for inspiring this study and providing helpful comments and ideas. This work was supported by STFC/UK Space Agency Grant ST/N000560/1. The authors declare that no competing financial interests exist.

7. References

Alfaro, A. C., F. Thomas, L. Sergent, and M. Duxbury (2006), Identification of trophic interactions within an estuarine food web (northern New Zealand) using fatty acid biomarkers and stable isotopes, *Estuar. Coast. Shelf Sci.*, *70*(1–2), 271–286, doi:10.1016/J.ECSS.2006.06.017.

Arthur, C. L., and J. Pawliszyn (1990), Solid Phase Microextraction with Thermal Desorption Using Fused Silica Optical Fibers, *Anal. Chem*, *62*, 2145–2148.

Benardini Iii, J. N., M. T. La Duc, R. A. Beaudet, and R. Koukol (2014a), Implementing Planetary Protection Measures on the Mars Science Laboratory, *Astrobiology*, *14*(1), 27–32, doi:10.1089/ast.2013.0989.

Benardini Iii, J. N., M. T. La Duc, D. Ballou, and R. Koukol (2014b), Implementing Planetary Protection on the Atlas V Fairing and Ground Systems Used to Launch the Mars Science Laboratory, *Astrobiology*, *14*(1), 33–41, doi:10.1089/ast.2013.1011.

Biller, P., A. B. Ross, and S. C. Skill (2015), Investigation of the presence of an aliphatic biopolymer in cyanobacteria: Implications for kerogen formation, *Org. Geochem.*, *81*, 64–69, doi:10.1016/J.ORGGEOCHEM.2015.01.010.

Blakkolb, B. et al. (2014), Organic cleanliness of the Mars Science Laboratory sample transfer chain, *Cit. Rev. Sci. Instruments J. Phys. Proc*, *85*(10),

480 75111–1043, doi:10.1063/1.1449727.

481 Borson, N. (2005), IEST-STD-CC1246D: product cleanliness levels and
482 contamination control program, *Clean Rooms*.

483 Botta, O., Z. Martins, C. Emmenegger, J. P. Dworkin, D. P. Glavin, R. P.
484 Harvey, R. Zenobi, J. L. Bada, and P. Ehrenfreund (2008), *Polycyclic*
485 *aromatic hydrocarbons and amino acids in meteorites and ice samples*
486 *from LaPaz Icefield, Antarctica*.

487 Brocks, J., E. Grosjean, and G. Logan (2008), Assessing biomarker syngeneity
488 using branched alkanes with quaternary carbon (BAQCs) and other
489 plastic contaminants, *Geochim. Cosmochim. Acta*, *72*(3), 871–888,
490 doi:10.1016/J.GCA.2007.11.028.

491 Calaway, M. J., C. C. Allen, and A. J. H (2014), *Organic Contamination*
492 *Baseline Study in NASA Johnson Space Center Astromaterials Curation*
493 *Laboratories*.

494 Cassidy, T., P. Coll, F. Raulin, R. W. Carlson, R. E. Johnson, M. J. Loeffler,
495 K. P. Hand, and R. A. Baragiola (2010), Radiolysis and Photolysis of Icy
496 Satellite Surfaces: Experiments and Theory, *Sp. Sci Rev*, *153*, 299–315,
497 doi:10.1007/s11214-009-9625-3.

498 Committee on preventing the forward contamination of Mars (2006),
499 *Preventing the Forward Contamination of Mars*, National Academies
500 Press, Washington, D.C.

501 Dachwald, B., J. Mikucki, S. Tulaczyk, I. Digel, C. Espe, M. Feldmann, G.
502 Francke, J. Kowalski, and C. Xu (2014), IceMole: a maneuverable probe
503 for clean in situ analysis and sampling of subsurface ice and subglacial
504 aquatic ecosystems, *Ann. Glaciol.*, *55*(65), 14–22,
505 doi:10.3189/2014AoG65A004.

506 Drake, M. J., D. S. Lauretta, and O. Team (2011), OSIRIS-REx Asteroid
507 Sample Return Mission, *Am. Geophys. Union, Fall Meet. 2011, Abstr.*
508 *#P42A-03*.

509 La Duc, M. T., R. Kern, and K. Venkateswaran (2004), Microbial Monitoring
510 of Spacecraft and Associated Environments, *Microb. Ecol.*, *47*(2), 150–
511 158, doi:10.1007/s00248-003-1012-0.

512 La Duc, M. T., S. Osman, P. Vaishampayan, Y. Piceno, G. Andersen, J. A.
513 Spry, and K. Venkateswaran (2009), Comprehensive census of bacteria in
514 clean rooms by using DNA microarray and cloning methods., *Appl.*
515 *Environ. Microbiol.*, *75*(20), 6559–67, doi:10.1128/AEM.01073-09.

516 Dworkin, J. P. et al. (2017), OSIRIS-REx Contamination Control Strategy and
517 Implementation,

518 Elsila, J. E., D. P. Glavin, and J. P. Dworkin (2009), Cometary glycine
519 detected in samples returned by Stardust, *Meteorit. & Planet. Sci.*,
520 *44*(9), 1323–1330, doi:10.1111/J.1945-5100.2009.TB01224.X.

521 Erd, C. (2012), Planetary Protection for the JUpiter ICy moons Explorer

(JUICE) Mission Candidate, *39th COSPAR Sci. Assem. Held 14-22 July 2012, Mysore, India. Abstr. B0.3-7-12, p.513*, *39*, 513.

Giger, W., and C. Schaffner (1978), Determination of polycyclic aromatic hydrocarbons in the environment by glass capillary gas chromatography, *Anal. Chem.*, *50*(2), 243–249, doi:10.1021/ac50024a020.

Grosjean, E., and G. Logan (2007), Incorporation of organic contaminants into geochemical samples and an assessment of potential sources: Examples from Geoscience Australia marine survey S282, *Org. Geochem.*, *38*(6), 853–869, doi:10.1016/J.ORGGEOCHEM.2006.12.013.

Haefliger, O. P., T. D. Bucheli, and R. Zenobi (2000), Laser Mass Spectrometric Analysis of Organic Atmospheric Aerosols. 1. Characterization of Emission Sources, *Environ. Sci. Technol*, *34*(11), 2178–2183, doi:10.1021/ES991122Y.

Harper, M. (2000), Sorbent trapping of volatile organic compounds from air, *J. Chromatogr. A*, *885*(1), 129–151, doi:10.1016/S0021-9673(00)00363-0.

Hauser, T. R., and J. N. Pattison (2019), *Analysis of Aliphatic Fraction of Air Particulate Matter*.

Hedgeland, R. J., P. A. Hansen, and D. W. Hughes (1994), Integrated approach for contamination control and verification for the Hubble Space Telescope first servicing mission</title>, in *Proc. SPIE 2261,*

543 *Optical System Contamination: Effects, Measurements, and Control IV,*
544 *(19 October 1994)*, vol. 2261, edited by A. P. M. Glassford, pp. 10–21,
545 International Society for Optics and Photonics.

546 Hodge, E. M., M. L. Diamond, B. E. McCarry, G. A. Stern, and P. A. Harper
547 (2003), Sticky Windows: Chemical and Biological Characteristics of the
548 Organic Film Derived from Particulate and Gas-Phase Air Contaminants
549 Found on an Urban Impervious Surface, *Arch. Environ. Contam.*
550 *Toxicol.*, *44*(4), 421–429, doi:10.1007/s00244-002-1272-6.

551 Illing, C. J., C. Hallmann, K. E. Miller, R. E. Summons, and H. Strauss
552 (2014), Airborne hydrocarbon contamination from laboratory
553 atmospheres, *Org. Geochem.*, *76*, 26–38,
554 doi:10.1016/J.ORGGEOCHEM.2014.07.006.

555 Johnson, P. V., R. Hodyss, V. F. Chernow, D. M. Lipscomb, and J. D. Goguen
556 (2012), Ultraviolet photolysis of amino acids on the surface of icy Solar
557 System bodies, *Icarus*, *221*(2), 800–805,
558 doi:10.1016/j.icarus.2012.09.005.

559 Kataoka, H., H. L. Lord, and J. Pawliszyn (2000), Applications of solid-phase
560 microextraction in food analysis, *J. Chromatogr. A*, *880*(1–2), 35–62,
561 doi:10.1016/S0021-9673(00)00309-5.

562 ten Kate, I. L., J. S. Canham, P. G. Conrad, T. Errigo, I. Katz, and P. R.
563 Mahaffy (2008), Mitigation of the Impact of Terrestrial Contamination on

564 Organic Measurements from the Mars Science Laboratory, *Astrobiology*,
565 *8*(3), 571–582, doi:10.1089/ast.2007.0160.

566 Kim, S.-K., and F. Karadeniz (2012), Biological Importance and Applications
567 of Squalene and Squalane, *Adv. Food Nutr. Res.*, *65*, 223–233,
568 doi:10.1016/B978-0-12-416003-3.00014-7.

569 Kimura, J., and N. Kitadai (2015), Polymerization of Building Blocks of Life
570 on Europa and Other Icy Moons, *Astrobiology*, *15*(6), 430–441,
571 doi:10.1089/ast.2015.1306.

572 Konstantinidis, K. et al. (2015), A lander mission to probe subglacial water on
573 Saturn′s moon Enceladus for life, *Acta Astronaut.*, *106*, 63–89,
574 doi:10.1016/j.actaastro.2014.09.012.

575 Lauretta, D. S. et al. (2015), The OSIRIS-REx target asteroid (101955) Bennu:
576 Constraints on its physical, geological, and dynamical nature from
577 astronomical observations, *Meteorit. Planet. Sci.*, *50*(4), 834–849,
578 doi:10.1111/maps.12353.

579 Lauretta, D. S. et al. (2017), OSIRIS-REx: Sample Return from Asteroid
580 (101955) Bennu, *Space Sci. Rev.*, *212*(1–2), 925–984,
581 doi:10.1007/s11214-017-0405-1.

582 Li, X., R. M. Danell, W. B. Brinckerhoff, V. T. Pinnick, F. van Amerom, R.
583 D. Arevalo, S. A. Getty, P. R. Mahaffy, H. Steininger, and F. Goesmann
584 (2015), Detection of Trace Organics in Mars Analog Samples Containing

585 Perchlorate by Laser Desorption/Ionization Mass Spectrometry,
586 *Astrobiology*, *15*(2), 104–110, doi:10.1089/ast.2014.1203.

587 Louch, D., S. Motlagh, and J. Pawliszyn (1992), Dynamics of organic
588 compound extraction from water using liquid-coated fused silica fibers,
589 *Anal. Chem.*, *64*(10), 1187–1199, doi:10.1021/ac00034a020.

590 Mahaffy, P. et al. (2003), *Report of the Organic Contamination Science*
591 *Steering Group*.

592 Martin, J. S. (1975), *Viking '75 program lander capsule sterilization plan*,
593 Hampton, Virginia.

594 Martins, Z., and M. A. Sephton (2010), Extraterrestrial Amino Acids, in
595 *Amino Acids, Peptides and Proteins in Organic Chemistry*, pp. 1–42,
596 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim, Germany.

597 McKay, C. P., C. C. Porco, T. Altheide, W. L. Davis, and T. A. Kral (2008),
598 The Possible Origin and Persistence of Life on Enceladus and Detection
599 of Biomarkers in the Plume, *Astrobiology*, *8*(5), 909–919,
600 doi:10.1089/ast.2008.0265.

601 Morris, H. C., L. A. Monaco, A. Steele, and N. Wainwright (2010), Setting a
602 Standard: The *Limulus* Amebocyte Lysate Assay and the Assessment of
603 Microbial Contamination on Spacecraft Surfaces, *Astrobiology*, *10*(8),
604 845–852, doi:10.1089/ast.2009.0446.

605 Nakamura, E. et al. (2012), Space environment of an asteroid preserved on

606 micrograins returned by the Hayabusa spacecraft, *Proc. Natl. Acad. Sci.*,
607 *109*(11), E624–E629, doi:10.1073/pnas.1116236109.

608 Neish, C. D., Á. Somogyi, and M. A. Smith (2010), Titan's Primordial Soup:
609 Formation of Amino Acids via Low-Temperature Hydrolysis of Tholins,
610 *Astrobiology*, *10*(3), 337–347, doi:10.1089/ast.2009.0402.

611 Nellen, J., P. Rettberg, G. Horneck, and W. R. Streit (2006), Planetary
612 protection – Approaching uncultivable microorganisms, *Adv. Sp. Res.*,
613 *38*(6), 1266–1270, doi:10.1016/j.asr.2005.10.026.

614 O'leary, W. M. (1962), The fatty acids of bacteria, *Bacteriol. Rev.*, *26*(4), 421–
615 47.

616 Otu, E. O., and J. Pawliszyn (1993), Mikrochimica Acta Solid Phase Micro-
617 Extraction of Metal Ions, *Mikrochim. Acta*, *112*, 41–46.

618 Pappalardo, R. T. et al. (2013), Science Potential from a Europa Lander,
619 *Astrobiology*, *13*(8), 740–773, doi:10.1089/ast.2013.1003.

620 Parkinson, C. D., M.-C. Liang, Y. L. Yung, and J. L. Kirschivnk (2008),
621 Habitability of Enceladus: Planetary Conditions for Life, *Orig. Life Evol.*
622 *Biosph.*, *38*(4), 355–369, doi:10.1007/s11084-008-9135-4.

623 Pflug, I. J. (1971), Planetary Quarantine. Sterilization of Space Hardware -
624 Journals - NCBI, *Environ. Biol. Med.*, *1*, 63–81.

625 Phillips, C. B., and R. T. Pappalardo (2014), Europa Clipper Mission Concept:

626 Exploring Jupiter's Ocean Moon, *Eos, Trans. Am. Geophys. Union*,
627 *95*(20), 165–167, doi:10.1002/2014EO200002.

628 Powell, J., J. Powell, G. Maise, and J. Paniagua (2005), NEMO: A mission to
629 search for and return to Earth possible life forms on Europa, *Acta*
630 *Astronaut.*, *57*(2), 579–593, doi:10.1016/j.actaastro.2005.04.003.

631 Probst, A. et al. (2012), Detecting inactivated endospores in fluorescence
632 microscopy using propidium monoazide, *Int. J. Astrobiol.*, *11*(02), 117–
633 123, doi:10.1017/S1473550411000425.

634 Reynolds, R. T., S. W. Squyres, D. S. Colburn, and C. P. McKay (1983), On
635 the habitability of Europa, *Icarus*, *56*(2), 246–254, doi:10.1016/0019-
636 1035(83)90037-4.

637 Sephton, M. A. (2002), Organic compounds in carbonaceous meteorites, *Nat.*
638 *Prod. Rep.*, *19*(3), 292–311, doi:10.1039/b103775g.

639 Sephton, M. A., J. H. Waite, and T. G. Brockwell (2018), How to Detect Life
640 on Icy Moons, *Astrobiology*, *18*(7), 843–855, doi:10.1089/ast.2017.1656.

641 Summons, R. E. et al. (2014), Planning Considerations Related to the Organic
642 Contamination of Martian Samples and Implications for the Mars 2020
643 Rover, *Astrobiology*, *14*(12), 969–1027, doi:10.1089/ast.2014.1244.

644 Tan, J., J. M. T. Lewis, and M. A. Sephton (2018), The Fate of Lipid
645 Biosignatures in a Mars-Analogue Sulfur Stream, *Sci. Rep.*, *8*(1), 7586,
646 doi:10.1038/s41598-018-25752-7.

647 Task Group on the Forward Contamination of Europa (2000), *Preventing the*
648 *Forward Contamination of Europa*, National Academies Press,
649 Washington, D.C.

650 Tielens, A. G. G. M. (2008), Interstellar Polycyclic Aromatic Hydrocarbon
651 Molecules*, *Annu. Rev. Astron. Astrophys.*, *46*(1), 289–337,
652 doi:10.1146/annurev.astro.46.060407.145211.

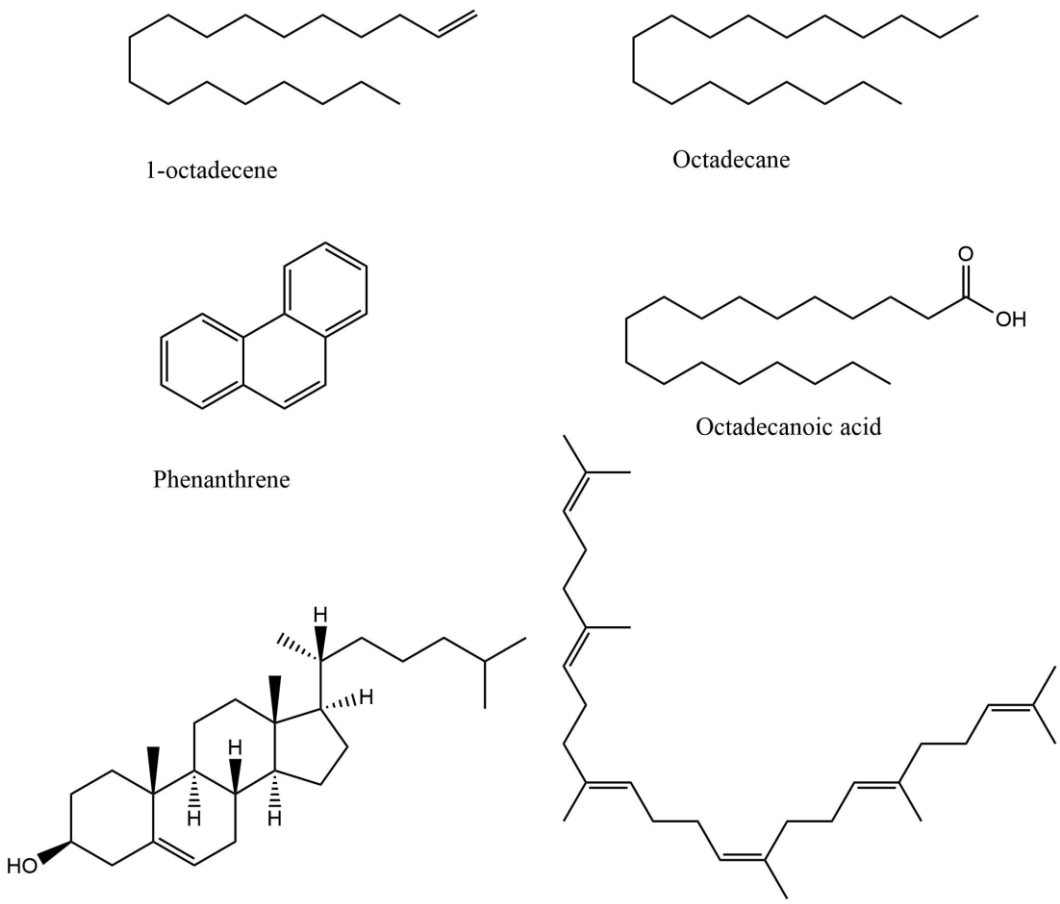
653 Trainer, M. G., A. A. Pavlov, J. L. Jimenez, C. P. McKay, D. R. Worsnop, O.
654 B. Toon, and M. A. Tolbert (2004), Chemical composition of Titan's
655 haze: Are PAHs present?, *Geophys. Res. Lett.*, *31*(17), n/a-n/a,
656 doi:10.1029/2004GL019859.

657 Tsuda, Y., M. Yoshikawa, M. Abe, H. Minamino, and S. Nakazawa (2013),
658 System design of the Hayabusa 2—Asteroid sample return mission to
659 1999 JU3, *Acta Astronaut.*, *91*, 356–362,
660 doi:10.1016/j.actaastro.2013.06.028.

661 Volkman, J. K., S. W. Jeffrey, P. D. Nichols, G. I. Rogers, and C. D. Garland
662 (1989), Fatty acid and lipid composition of 10 species of microalgae used
663 in mariculture, *J. Exp. Mar. Bio. Ecol.*, *128*(3), 219–240,
664 doi:10.1016/0022-0981(89)90029-4.

665 Westphal, A. J. et al. (2014), Final reports of the Stardust Interstellar
666 Preliminary Examination, *Meteorit. Planet. Sci.*, *49*(9), 1720–1733,
667 doi:10.1111/maps.12221.

668



669

670 *Figure 1 Standard organic compounds selected for SPME organic contamination control method*
671 *development*

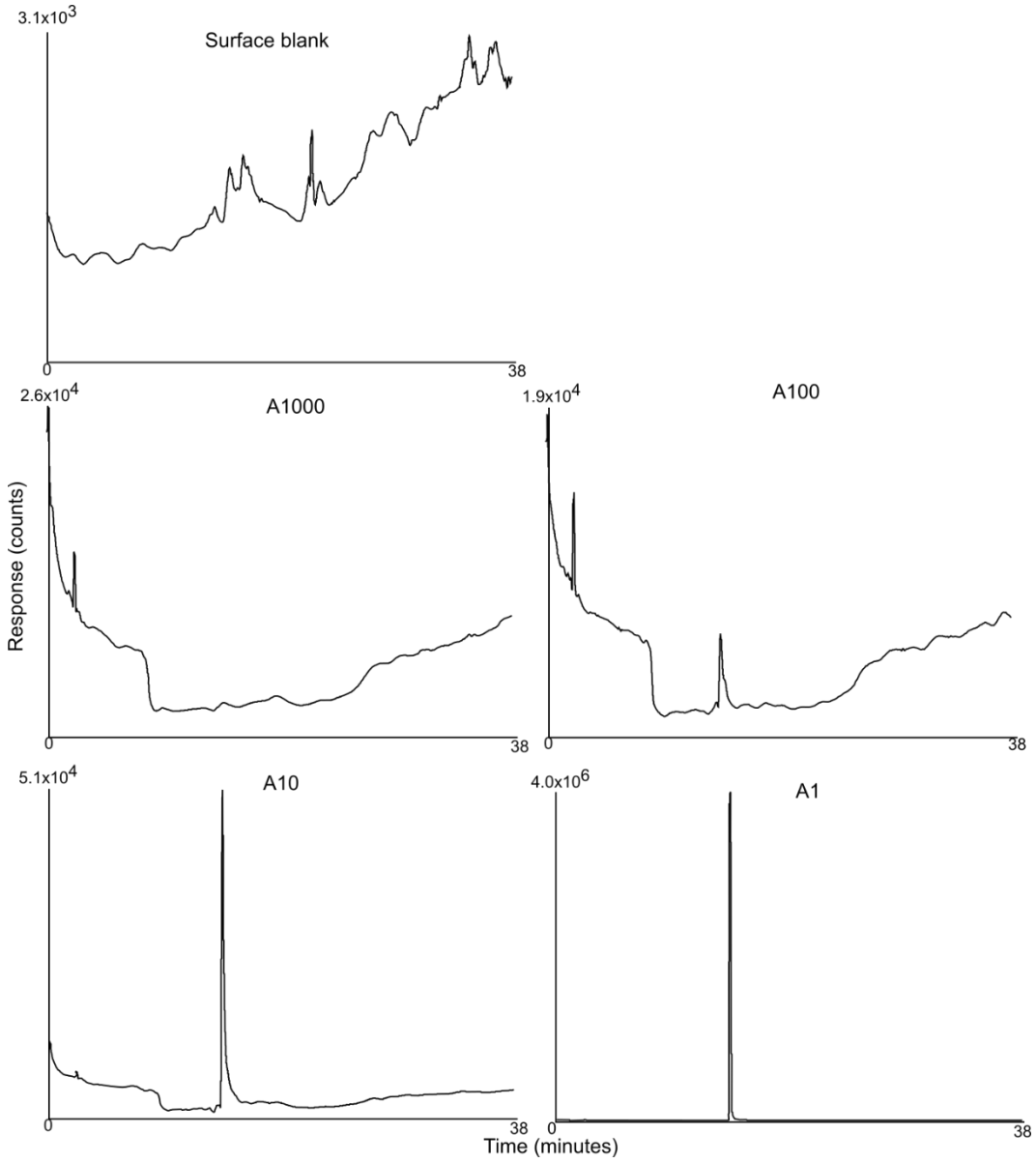


Figure 2 SPME-GC-MS mass 57 chromatograms of octadecane spiked onto test surface at varying concentrations. Octadecane peak is at ~16 min 40 s, below LOD at A1000 contamination level, although a small peak is visible.

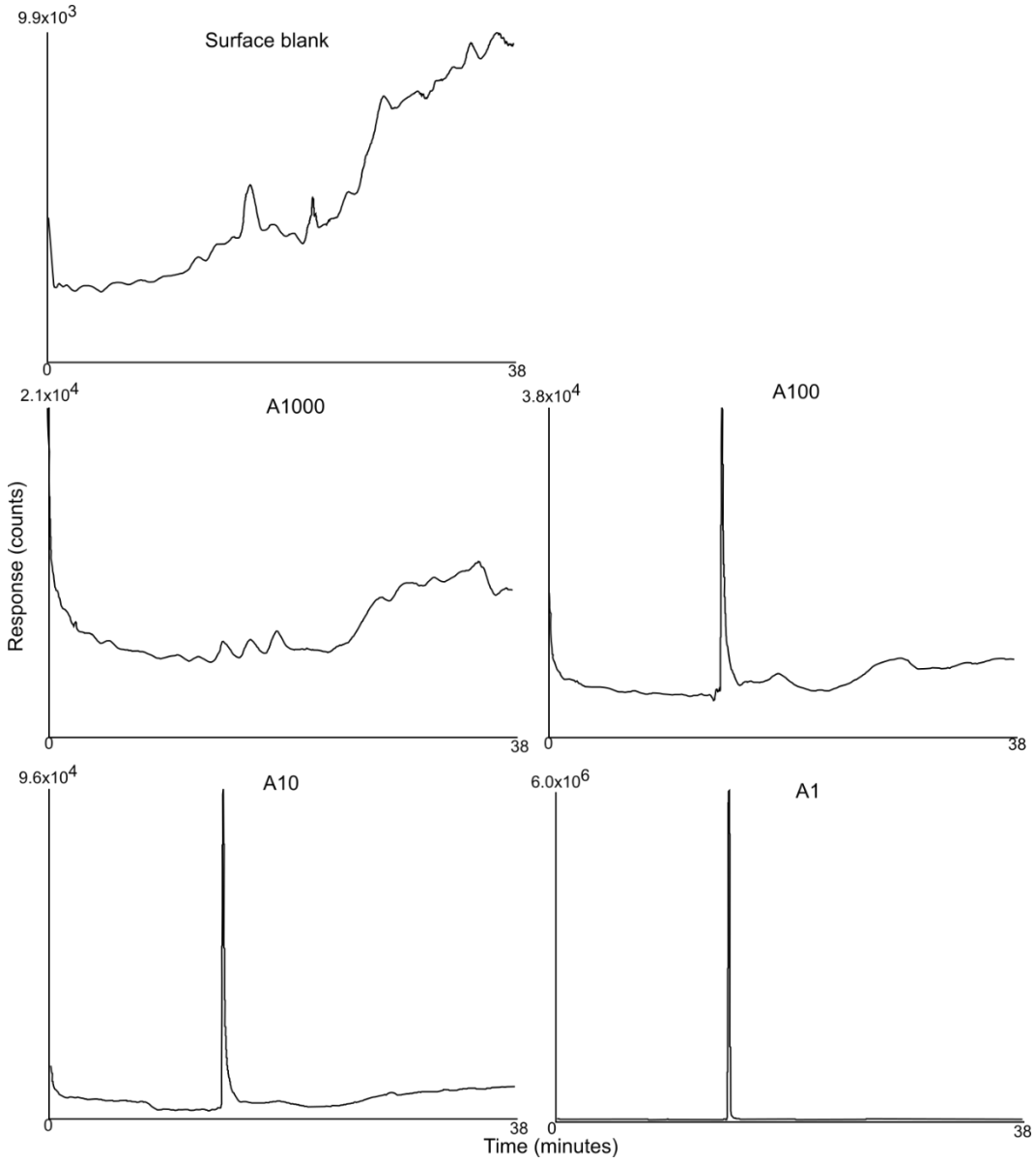


Figure 3 SPME-GC-MS mass 55 chromatograms of 1-octadecene spiked onto test surface at varying concentrations. 1-octadecene peak is at ~16 min 30 s, null detection at A1000 contamination level.

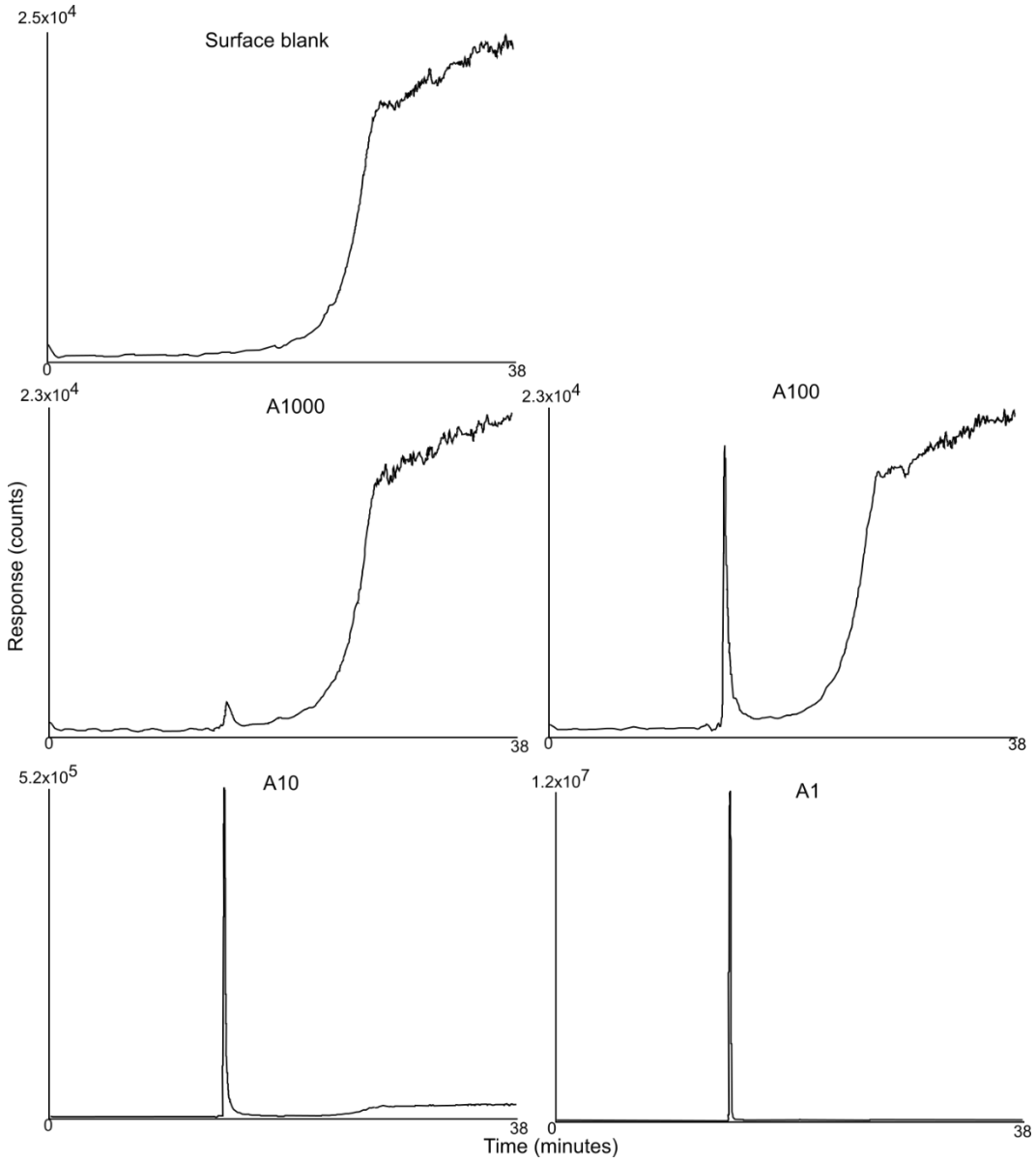


Figure 4 SPME-GC-MS mass 178 chromatograms of phenanthrene spiked onto test surface at varying concentrations. Phenanthrene peak is at ~16 min 40 s.

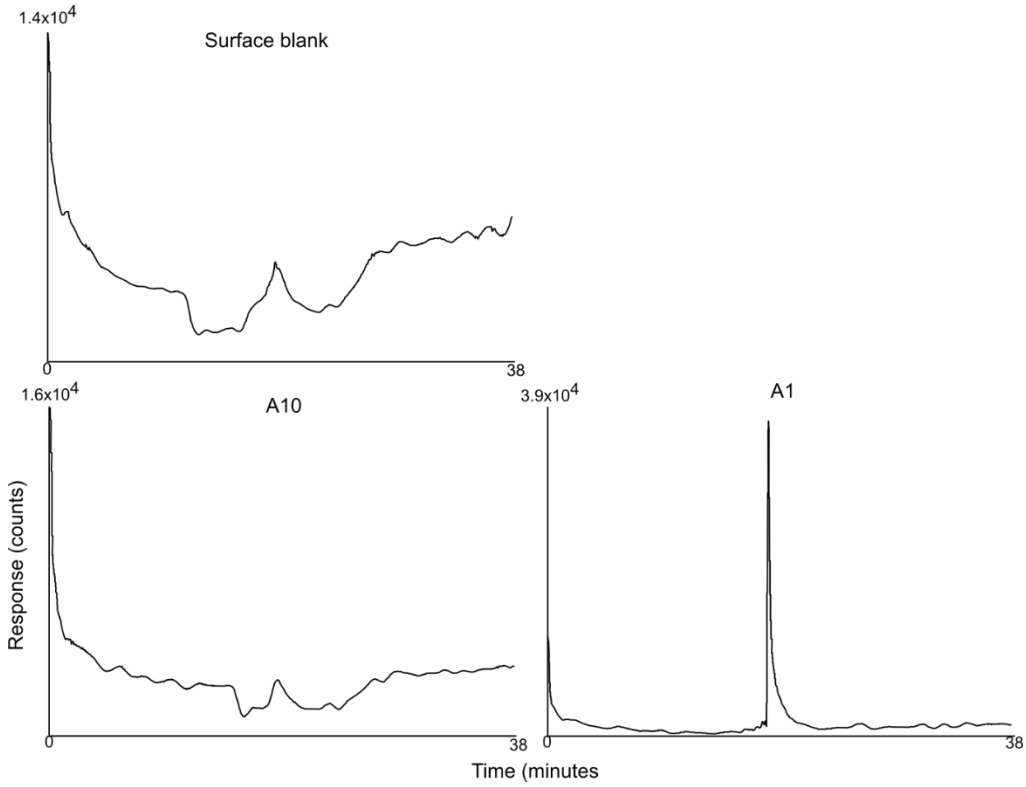


Figure 5 SPME-GC-MS mass 57 chromatograms of octadecanoic acid spiked onto test surface at varying concentrations, experiments at concentrations equal to A1000 and A100 were not run as the liquid injection experiments showed the GC-MS method to be insufficiently sensitive at these concentrations. Octadecanoic acid peak is at ~20 min 10 s, below LOD at A1000 contamination level, background peak at 20 min 40 sec appears in blank and may obscure small octadecenoic acid peak.

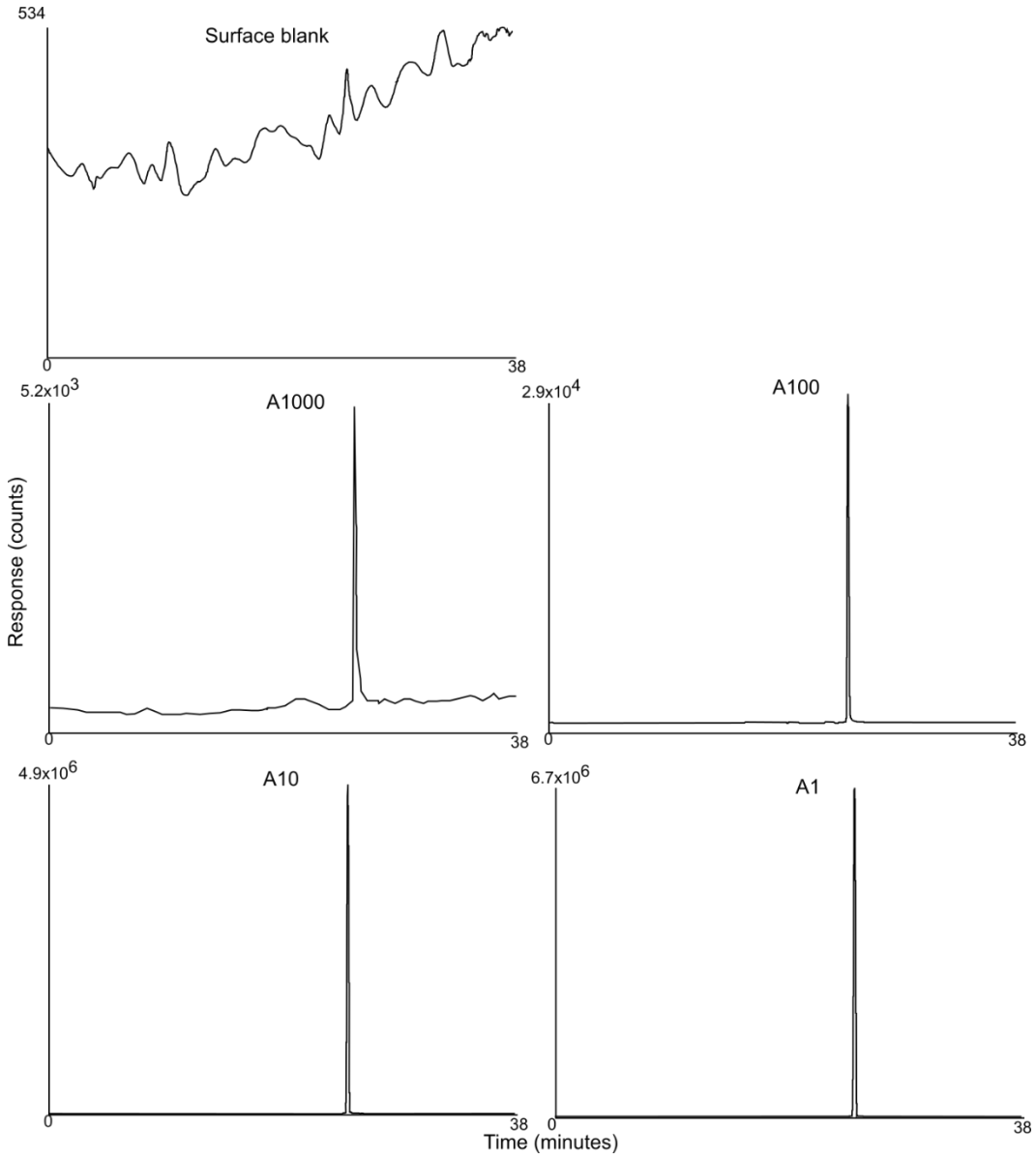
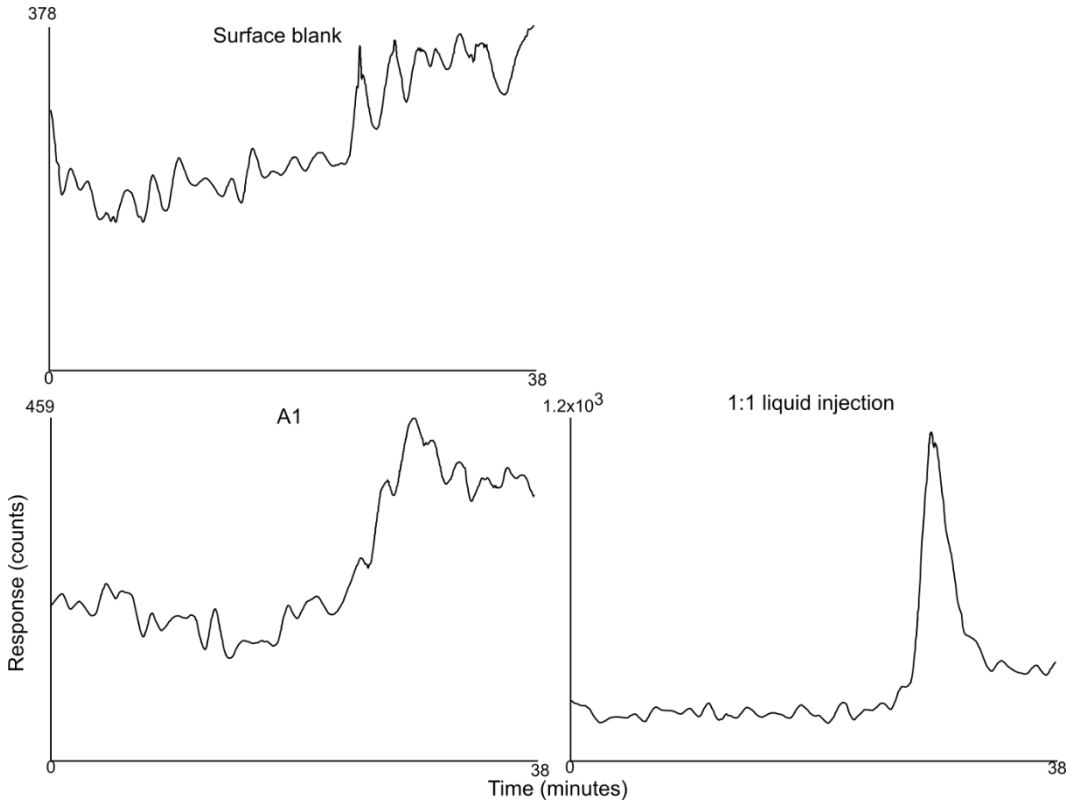


Figure 6 SPME-GC-MS mass 69 chromatograms of squalene spiked onto test surface at varying concentrations. Squalene peak is at ~25 min 45 s.



692

693 *Figure 7 SPME-GC-MS mass 217 chromatograms of cholesterol spiked onto test surface at varying*
694 *concentrations, experiments at concentrations equal to A1000, A100, A10 were not run as the liquid*
695 *injection experiments showed the GCMS method to be insufficiently sensitive at these concentrations.*
696 *Cholesterol peak is at ~29 min 20 s , this is below LOD even at A1 contamination level, although a small*
697 *broad hump is observed corresponding with this retention time. The positive detection of cholesterol in*
698 *the 1:1 (mg to ml) liquid injected sample is shown for comparison.*

699

700 *Table 1 Contaminants of concern for astrobiological missions [after Mahaffy et al., 2003]*

Molecular class	**Examples**
Aromatic hydrocarbons	Benzene, tolulene, higher molecular weight aromatics, polyaromatic hydrocarbons
S, N, O heterocyclic aromatics	Furan, pyridine, pyramadine, benzothiophene
Carboxylic acids & their salts	Alkyl & aromatic acids, fatty acids
Aliphatic hydrocarbons	Alkenes, alkanes
Nitrogen containing compounds	Amino acids, amines, amides, purines, pyrmidines, porphyrins
Alcohols	Methanol, higher molecular weight linear and branched chain alcohols
Carbonyl	Esters, ketones, aldehydes
Sulfonic, phosphonic acids	Methanesulfonic acid
Lipids and derivatives	Hydrocarbon chains, fatty acids, fats, phospholipids, hopanes, steranes
Sugars and derivatives	Glucose
Proteins	Polar and non-polar
Nucleotides	DNA fragment

701

702

703 *Table 2 List of methods and their sensitivities suggested for testing contamination levels on the Mars*
704 *2020 rover, adapted from Summons et al [2014]*

	Method	Sensitivity	Sampling/Form*	Comments
Vibrational Spectroscopy	DRIFT spectroscopy	<1 ng/cm^2 (from 100 cm^2)	Witness plate or solvent extract**	Provides broad range of chemical functional groups and/or identification. Applied to numerous spacecraft mission, detects common airborne contaminants (AC) and spacecraft molecular contamination. Large spacecraft database.
	FTIR-Grazing angle attenuated total reflection IR (GATR)	Sub-monolayer 0.5 ng/cm^2	Witness plate or solvent extract	Provides chemical functional groups and identification, detects common AC. Rapid
	FTIR-Microscopy	Sub-nanogram particles	Specialized witness plate	Requires specialized witness plates or particle sampling. Rapid.
	Raman-Microprobe	Sub-nanogram particles	Specialized witness plate	Requires specialized witness plates or particle sampling. Rapid.
Mass Spectrometry (MS)	GC-MS	<0.1 ng/cm^2 (from 100 cm^2)	Witness plate or solvent extract	Identification of components in a complex mixture. Non-volatile components not detected, detects common AC
	Pyrolysis GC-MS	<0.1 ng/cm^2 (from 100 cm^2)	Witness plate or solvent extract	Detects non-volatile components, can run in series with GC-MS.
	Direct Analysis in Real Time (DART)-MS	<0.001 ng/cm^2 (from 100 cm^2)	Witness plate or solvent extract	Identification of components in a complex mixture, molecular weight >1000 amu requires pyrolysis, detects common AC, very sensitive, rapid
	Liquid Chromatography (LC)-MS	<0.1 ng/cm^2 (from 100 cm^2)	Witness plate or solvent extract	Identification of components in a complex mixture, somewhat complex procedures and method development, particularly well-suited for some biological analyses.
	Laser-assisted Desorption (LD)-MS	<1 ng/cm^2	Witness plate or solvent extract	Identification of components in a complex mixture, suited for high molecular weight bio-analytes, complex procedures and method development, expensive instrumentation.
	Secondary-Ion MS (SIMS)	Sub-monolayer	Witness plate	Quantitation difficult, limited molecular identification for organics, very sensitive, detects common AC, complex, expensive instrumentation
Other	X-ray Photoelectron Spectroscopy (XPS)/Auger	Sub-monolayer	Witness plate	Sensitive, elemental information, limited molecular identification, detects common AC, complex, expensive instrumentation
	Total Organic Carbon (TOC) Instruments (pyrolysis and electrochemical)	~3 ng/cm^2	Witness plate	No chemical information, no identification, does not quantify incombustible components

705 **It should be noted that all methods require specialized hardware sampling and/or witness plates*
706 ***Solvent extracts may use a surface rinse or specialized solvent swabs of hardware surfaces*
707

708

Table 3 Potential organic contaminants and the Supelco recommended fibres for their analysis (by headspace extraction) (adapted from https://www.sigmaaldrich.com/technical-documents/articles/analytical/selecting-spme-fibers.html#fiber)

Analyte type	**Recommended Fibre**
Low molecular weight	75 µm/85 µm Carboxen/Polydimethylsiloxane
Volatiles	100 µm polydimethylsiloxane
Volatile, amines and nitro-aromatics	65 µm polydimethylsiloxane/divinylbenzene
Polar semi-volatiles	85 µm polyacrylate
Non-polar high molecular weight compounds	7 µm polydimethylsiloxane
Non-polar semi-volatiles	**30 µm polydimethylsiloxane**
Alcohols and polar compounds	60 µm Carbowax

Table 4 Integrated peak areas from selected ion chromatograms produced by both liquid injection and SPME-GC-MS of standard organic compound solutions. Percentage recovery of standard compounds against liquid injection external standard calibration (N/A = Not applicable, <LOD = below limit of detection)

	Solution concentration (ng: ml)	1 µl direct liquid injection (peak area)	Contamination level equivalent	SPME average (peak area)	SPME Standard deviation (% of mean)	% recovery at contamination level
Octadecane	0.1	<LOD	N/A	N/A	N/A	N/A
	1	941	A1000	<LOD	N/A	<LOD
	10	5207	A100	1723	24 %	9 %
	100	61663	A10	8694	50 %	5 %
	1000	1859793	A	368767	63 %	20 %
1-Octadecenee	0.1	<LOD	N/A	N/A	N/A	N/A
	1	1688	A1000	<LOD	N/A	<LOD
	10	4677	A100	9411	18 %	57 %
	100	90213	A10	20120	14 %	12 %
	1000	1650655	A	913332	48 %	55 %
Phenanthrene	0.1	577	N/A	N/A	N/A	N/A
	1	3309	A1000	1379	8 %	25 %
	10	23632	A100	6227	23 %	11 %
	100	434883	A10	101058	34 %	18 %
	1000	5510655	A	1761618	18 %	32 %
Octadecanoic acid	0.1	<LOD	N/A	N/A	N/A	N/A
	1	<LOD	A1000	N/A	N/A	<LOD
	10	<LOD	A100	N/A	N/A	<LOD
	100	11345	A10	<LOD	N/A	<LOD
	1000	549876	A	14439	31 %	3 %
Squalene	0.1	85	N/A	N/A	N/A	N/A
	1	336	A1000	611	20 %	99 %
	10	4388	A100	4030	9 %	65 %
	100	72063	A10	48410	19 %	78 %
	1000	653790	A	509926	67 %	82 %
Cholesterol	0.1	<LOD	N/A	N/A	N/A	N/A
	1	<LOD	A1000	N/A	N/A	<LOD
	10	<LOD	A100	N/A	N/A	<LOD
	100	<LOD	A10	N/A	N/A	<LOD
	1000	863	A	<LOD	N/A	<LOD

Table 5 Calculated sensitivities for each standard compound and concentration (expressed as percentages)

Level	Octadecane	1-Octadecene	Phenanthrene	Steric acid	Cholesterol	Squalene
A	100	100	100	100	0	100
A10	100	100	100	0	0	100
A100	100	100	100	0	0	100
A1000	0	0	100	0	0	100

725 *Table 6 molecular weights and enthalpy of vaporization of standard compounds tested (from NIST)*

Standard compound	Molecular weight	Enthalpy of vaporization (KJ/mol)
1-Octadecene	252	55
Octadecane	255	92
Phenanthrene	178	78
Steric acid	284	79
Cholesterol	387	154
Squalene	410	83

726

727

728