Much of astrobiology relies on the use of analog sites as a means to understand extreme environments on other planets. We are interested in understanding anthropogenic effects on the Mojave Desert microbial communities and chemical environment. To do this, we ask, are microbial communities, as well as the areas chemical and environmental dynamics, different at various locations along I-40s desert corridor?. Using a custom Deep Ultraviolet Native Fluorescence/Raman Spectrometer (DU-VNF/RS), we are non-invasively detecting the types of organics and microbes deposited on samples exposed to the Mojave environment. We placed sterile/clean inorganic surfaces at three locations in the Mojave for up to three months. Two locations were adjacent to the I-40, and another at a more remote location. Samples were collected and scanned by deep UV native florescence to generate 2D chemical maps. The fluorescence maps showed both organic and inorganic deposition with chemical variability between samples based on location. Secondary analysis with deep UV Raman provided additional detail. In addition to organic analysis, the totality of the number of samples and amount of data collected led to the development of a new custom database system to organize results and ease data analysis.

Introduction

Astrobiology uses sites on Earth with similar conditions to those found on planets and moons within our solar system. In particular, we are interested in identifying sites as analogs for Mars. The Mojave Desert has similar physical conditions and chemical composition to the red planet and is often used as a mechanical analog for testing rovers [Marlow, J. J., et al., 2008]. The basaltic characteristics of its rock are of keen interest to astrobiologists and are used often in the laboratory to simulate martian dust [Peters, G. H., et al., 2008]. The Mojave also has the benefit of close proximity to the Jet Propulsion Laboratory.

Characterizing the level of contamination at these sites is a priority if we want to successfully approximate Mars conditions on Earth. The Atacama Desert is a very popular analog for Mars that is used to study habitability because of its similar soil conditions and its scarcity of organics [Marlow, J. J., et al., 2008]. Yet, even here, human activity and industrial traffic are not too far away. Therefore, it is imperative that, when using analogs to investigate the possibility of
finding life elsewhere, we must first discern which of the organics present are indigenous and which are due to contamination.

Despite the popularity of the Atacama and Mojave sites and several others around the planet, little work has been done to determine the level of contamination by anthropogenically caused organic contamination (common contaminations include diesel exhaust from trucks, tarring of the road, mining sites, etc). How far away from human activity must we get before we see a drop off in pollution? We consider our investigation of the Mojave as a first step in developing methodology for selecting future analogs.

Methods

We chose two sites, one in close proximity to human activity and the other away that could be used as a control. Since I-40 is a major corridor through the Mojave Desert and a major source of anthropogenic organic contamination, we selected a site near the Pisgah Crater volcanic fields. Pisgah contains basaltic lava flow and is within 50 meters of the freeway. We selected sites on both the north and south sides of the freeway since we were also interested in testing how wind direction might affect deposition of organics coming from passing traffic. The second location was near Amboy Crater which has similar volcanic rock compositions as those found at Pisgah. Amboy is approximately 40 km from the nearest major highway (I-40).

Because of the Mojaves popularity as a martian analog, we selected mineralogy similar to that found on Mars. This included calcite, sandstone, and basalt. Stainless steel coupons were also used in the experiment. We secured the individual pieces of rock and steel using a putty glue. We also labeled the substrates and noted the date, time, and direction of their placement for later reference. We duplicated these procedures at Amboy Crater.

CO2 and temperature monitors were also left with the samples. We also collected data from the local Barstow-Daggett airport weather station. We developed weather analysis software (METAR) to track various weather patterns that could be graphed for comparison. In particular, METAR was valuable for tracking wind trends using a customized wind rose chart.

After approximately a month of deposition, the stainless steel coupons were removed and replaced.
with a fresh set in mid-March 2013. The second set was left for a two month period. The coupons and mineral substrate samples were removed in mid-May 2013 and brought into the lab for analysis.

DUVNF/RS Instruments

We analyzed our samples using the latest in deep UV native fluorescence and Raman spectroscopy (DUVNF/RS) technology. Our DUVNF/RS instruments are novel devices built on lab at JPL. The instruments are able to characterize biology due to the presence of aromatic compounds, in particular aromatic amino acids found in microbial proteins. Using our instruments, we are able to discriminate among bacterial cells, spores, and organics. [Bhartia, R., et al., 2008] Deep UV spectroscopy (<250nm) also has the added benefit of separating the fluorescence from the Raman region. [Asher, S.A. and C.R. Johnson, 1984] Furthermore, the use of deep UV Raman spectroscopy has gained recognition in recent years as a valuable method of planetary in-situ investigation [Tarcea, N., et al., 2007].

MOSAIC and MOBIUS are both DUVNF/RS capable. However, MOSAIC is currently set up to produce two dimensional native fluorescence chemical mapping while MOBIUS is designed to do both DUVNF/RS point spectral mapping. The benefits of this type of technology are not well-documented and this experiment was helpful in demonstrating its potential.

Each sample returned from the field was initially run under MOSAIC. We used a 250-micrometer laser spot size. The instrument moves across the entire samples surface and collects spectral data over the designated spot size. The software then combines...
Figure 3: Our initial collection included calcite, sandstone, basalt, and stainless steel coupons.

Figure 5: MOSAIC instrument.

Figure 6: MOBIUS instrument.

all of the data into a single file. The spectrometer uses an ultraviolet laser that produces a 248.6nm wavelength. MOSAIC contains six channels at varying wavelengths that are calibrated, incrementally, to sensitivities between 280 and 380nm with a difference of 20nm between each channel. The rationale for this is that the light that returns from the sample will be at a lower energy than the light originating from the laser.

Furthermore, unlike in other techniques, the use of a spot size that is greater than a micron facilitates easier characterization of microbes on both man-made and natural surfaces with larger surface areas in real-time [Asher, S.A. and C.R. Johnson, 1984]. On average, samples with approximately 25 cm area required less than 30 minutes for map acquisition.

We extracted seven maps in total for each sample using custom map constructor software. One map
Figure 7: Two-dimensional spectra maps of a stainless steel plate shows possible organic deposition. We can see variation in deposition by extracting images at each band for which MOSAIC detects the returning light.

included all six channels (or bands) of the instrument while the remaining six maps were extracted as individual bands. Extracting the individual bands allows for better analysis of the mapped surface.

Samples that showed regions with organic material were further analyzed using Raman spectroscopy and the MOBIUS instrument.

With MOBIUS, we were able to zoom into a 200 wide micron section of one of the stainless steel coupons from Pisgah. We took several point spectra in regions that showed possible high microbial distribution.

Results and Discussion

During the first collection period, we noticed that the putty's oils were seeping into the rock substrates. We later confirmed the presence of these oils using deep UV native fluorescence mapping. The maps were oversaturated and forced us to limit our analysis to the stainless steel coupons which are far less porous. In total, we were able to use 12 stainless steel coupons, four from each site, in our analysis.

We have four preliminary results. First, we were able to identify deposition variation over time. Our two-dimensional maps showed that the first set of samples had less organic deposition than the second set, which were left out for a longer period of time (approximately two months). We also noticed that Pisgah had greater deposition in comparison to Amboy. This was expected and indicates that our use of Amboy as a control for the experiment was well warranted.

Second, we noticed that environmental dynamics played a role in the way organics are deposited onto the substrate. The steel coupons from one of the Pisgah sites show greater deposition and suggest a southerly wind direction which matched the data graphed by our wind rose software.

Third, as mentioned previously, the coupons left at Amboy crater showed less organic deposition than the ones left at Pisgah. This was consistent regardless of the duration that samples were left out. This suggests that when scouting for analogs, some kilometer distance from intense human activity is vital, especially when using sites for chemical analysis. The Amboy site was approximately 40 km from Interstate 40 and approximately 1 km from a lightly used road and main line railroad tracks.

Lastly, our results showed signs of microbial deposition alongside abiotic organics. Initial analysis of the deep UV fluorescence two-dimensional maps showed large regions of organic deposition on coupons that
Figure 10: Side by side comparison of stainless steel plates. Plate A from the Amboy site shows little deposition of organics while plate B shows areas of organic deposition as indicated by the arrows.

were left out for a two-month period. The maps are limited, though, and provided only an overview of the surface deposition. They cannot show the types of organics that collected on the coupons. We used Raman spectroscopy to determine the types of organics present.

Our Raman spectral analysis showed signs of an unknown biomass in the region of interest on a Pisgah stainless steel coupon. Peak signatures showed a mixture of organics that are consistent with a microbial signature described in Manoharan et al. 1991 where they did deep UV Raman spectroscopy of microbes [Manoharan, R., et al., 1991]. Detailed analysis is ongoing to better understand the nature of the microbe whether it is yeast, bacterial, or fungal. This can potentially be determined using the Raman spectra we have taken with MOBIUS. This depends upon further analysis, peak fitting, smoothing, and determining which spectra that are already identified and available match our data. Within the 700 micrometer region, we also found areas that mainly showed spectra typical of silica or hematite. The silica is likely a result of dust that collected on the coupon and hematite a likely result of the steel oxidizing.

Since this was a pilot study designed to determine the requirements for evaluating potential new analog sites, we expected some problems with our initial approach. From some of the less than successful approaches, we can report that using putty to keep samples on the rocks is inadvisable. In order for our results to be more definitive, we would need to find a better way to keep the samples in place without contaminating the substrates. In an unrelated study performed in our lab at JPL, we found that samples left out in the laboratory are not protected from contamination. We would like to be able to run the experiment in-situ to avoid such unwanted contamination. Furthermore, we would like to develop a better technique to identify which substrates certain contaminants might prefer.

We would also like to gather more samples from the environment to see what general trends we might find in the way deposition occurs. Do wind patterns vary significantly from one location to another? We saw some interesting striations on some of the plates that do not have an immediate explanation. The collection of more data, however, led us to the realization that a database was needed to make analysis more meaningful.

As a result of this realization, we developed a web-based database called MIND (Multi-INstrument Database). The database was built using the latest web development languages, including PHP, JavaScript, HTML, CSS, and MySQL. The database
can be accessed from any web browser. Samples can be entered and associated with the various instruments in the lab, or projects, for example. Sample entries can also be updated and searched.

This experiment was part a year-long evaluation of techniques designed to prepare for an instrument being proposed for the Mars 2020 payload. The deep UV instrument called SHERLOC (Scanning Habitable Environments with Raman Luminescence for Organics and Chemicals) is currently being proposed as an arm instrument for in-situ, non-contact, non-invasive detection and distribution of organics and minerals; much of the technology in SHERLOC leverages off of MOBIUS and MOSAIC. As the data collection and analysis during this study progressed, it became clear that a database to organize the data from multiple instruments was necessary. MIND is being designed not just to house data from this experiment, but to house data over many, many experiments.

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References


