**The Astrobiology Field Laboratory**

September 26, 2006

Final report of the MEPAG Astrobiology Field Laboratory Science Steering Group (AFL-SSG)

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During the course of the SSG several breakout groups were formed to answer specific issues related to our discussions. These are as follows;

AFL subcommittees

**Sedimentary sub-team**. Pan Conrad, leader.

**Hydrothermal sub-team**. David Blake, leader

**Ice sub-team**. Luther Beegle, leader

**Sample preparation sub-team**. Jan Toporski, leader

**Definitions sub-team**. Pan Conrad, leader

**Instruments sub team.** Will Brinkerhoff leader

**Water sub-team**. Jan Amend, leader

# EXECUTIVE SUMMARY

The AFL SSG was asked to develop an analysis of a possible future mission called the **Astrobiology Field Lab**. This mission is a generic concept, consisting of a lander equipped with a major in-situ laboratory capable of making significant advancements towards MEPAG’s Goal I (“Determine if life ever arose on Mars”). In essence, the purpose of this analysis was to evaluate the question, “what is the most that can be accomplished in this area by in situ means?” In order to give the analysis team room to work, financial and timing constraints were very loose. Although at the time of convening this exercise 2013 was the closest discussed deadline and so considerations were given to what technically could be accomplished for this deadline.

The AFL SSG considered the problem at several levels:

What overall programmatic exploration strategies are needed to achieve Goal I? Results from many missions will contribute to these strategies, and a mixture of ambiguous and definitive outcomes will need to be accommodated.

What result would AFL need to deliver to make a meaningful contribution to this strategy?

What are the engineering options for configuring a landed mission that would make such a contribution?

Programmatic exploration strategies

In order to plan missions during the period 2013-1018, it is necessary to predict the state of human knowledge at that time. Although this is hard to do in detail, it is possible to reach some important generalities. First of all, habitability is the potential of an environment (and applied to either the past or the present) to sustain life. By this definition, habitability will be the integrated and accumulated knowledge of many missions and many different kinds of scientific investigations. However, as with any other potential, it will not be possible to achieve certainty unless life itself is discovered. Habitation, on the other hand, is a simple yes-no question. A key planning question, therefore, is when has the habitability potential risen high enough that a habitation test can be justified?

Although it has been generally assumed in the past that these two objectives need to be pursued sequentially, the AFL SSG has concluded that organisms and their environment together constitute a system, and each produces an effect on the other. Many kinds of investigations of this system can simultaneously provide information about both. This implies that habitability and habitation can be investigated together. This expands significantly on the current mission concept for MSL, with AFL having an expanded instrument suite dedicated more towards life detection and precision sample handling than MSL. Moreover, the process of life detection on Mars involves two sequential steps: 1). Proposing that a set of phenomenon are, or could be, biosignatures. This will constitute a working hypothesis that life is or was present. 2). Establishing that at least one of these biosignatures is definitive. This requires extensive effort and careful planning and a number measurements mutually confirming each other. Finally, we know that some kinds of scientific investigations will measure signs of both extinct and extant life without needing to distinguish between these two possibilities before launch.

Given the expected state of our knowledge about Mars during the period 2013-2018, the AFL SSG has reached three conclusions:

It is both possible and reasonable to do life detection first, then determine whether it is extinct or extant on the basis of a positive result.

Missions during this period can reasonably begin the process of life detection by characterizing potential biosignatures.

It is reasonable to set mission objectives that relate to both habitability AND habitation. It is not necessary to choose one at the expense of the other.

Finally if a definitive biosignature is located by AFL instrumentation and missions must be configured to definitively characterize that life signature. It is only by thorough study of a positive signal will skepticism be kept to a minimum and the maximum understanding of how this relates to the formation of life on earth be understood.

Engineering options

The AFL SSG has concluded that the following overall scientific objective is both achievable by AFL as early as 2013 (although 2018 was also postulated as a target from the pathways document, Figure 1), and is a significant extension of currently planned missions:

**For at least one Martian environment of high habitability potential, quantitatively investigate the geological and geochemical context, the presence of the chemical precursors of life, and the preservation potential for biosignatures, and begin/continue the process of life detection.**

By targeting an environment of high habitability potential, a response to prior discoveries is implied. Investigating the context is a reflection of the reality that our understanding of habitability will not be complete by 2013 we need to plan for more work. Understanding prebiotic chemistry is necessary to allow planetary-scale life-related predictions, especially in the contingency that life is not found in a specific experiment. Understanding preservation is key to interpreting the results of biosignature investigations, and is also critical feed-forward to future missions. Finally, life detection, as AFL SSG defines it, is a process that will take time. It is reasonable to expect that missions like AFL will play a significant role in this process, but unreasonable to expect that they will bring it to a conclusion.

Engineering options for an AFL mission

The AFL SSG has defined a landed mission that can achieve the above objective. There are multiple possible variations of what could be called “AFL”, and different scientists see these variations in different context, and with different systems of priority. However, it is possible to define an invariant base that is common to most versions, along with a discovery-responsive and competition-responsive cap. The basic landed system needs to be able to accomplish four things:

Acquire the right samples (access a place with high general habitability potential, understand preservation potential, have a high ability for scientific sample selection, capable sample acquisition system)

Know the context (Setting, mineralogy, chemistry, relationships)

ID best place on the sample (Mid-scale observations.

Precision sub-sampling (down to mm scale) for investigation by analytical suite)

At least 3 mutually confirming A/B measurements (Suites of observations by different means of the same or related phenomena will be necessary to reach definitive conclusions).

Initial engineering concepts for this mission place AFL as a COSPAR level 4B mission.

# 2.0AFL CHARTER

The AFL SSG was given the following charter.

Introduction

The Mars Program Office at NASA HQ (Code S) requests a study of the preliminary scientific options and engineering characteristics of the AFL mission. This mission was identified in the final report of the MSPSG (Mars Science Program Synthesis Group).

Starting assumptions (to be refined)

Assumptions for each mission need to be compiled separately.

Assume TBD mission must be ready to launch as early as TBD.

Science priorities will be derived from the MEPAG Goals document.

Requested Tasks:

Develop a set of candidate whole mission concepts. For each:

Define preliminary general science objectives, and science floor (the level below which the mission is not worth flying).

Identify and evaluate the primary science trades

Determine whether instruments capable of addressing the science objectives are likely to be available in time.

Landing site accessibility: Propose the size of the latitude band which needs to be held open for this mission, the landing precision, and required ability to land in rough terrain

Identify possible facility subsystems related to sample acquisition and sample preparation.

Describe the essential engineering constraints on the mission

Determine if positioning in the pathways makes a difference to the science/engineering of the mission.

Describe how the mission fits into NASA’s long-range strategic framework for the exploration of Mars

Based on the above analysis, present a prioritized set of preliminary options for consideration by NASA HQ.

Methods

The SSG is asked to conduct its business primarily by telecons, e-mail, and or web-based processes. There is enough budget to convene 1 or 2 face-to-face meetings.

Logistical support will be provided by the Mars Program Science Office.

Timing

It is expected that the team will be ready to start its deliberations in mid-November.

A mid-term telecon status check by Jim Garvin, Dan McCleese, and Bruce Jakosky is requested after the new year.

The near-final report of the AFL SSG is requested by Feb. 28, 2004.

It is expected that the results of this study will be presented to MEPAG at its June, 2004 meeting. Feedback from this discussion will be incorporated in the final report, which will be due July 31, 2004.

Report Format

It is requested that the results be presented in the form of both a PowerPoint presentation and a white paper. Additional supporting documents can be prepared as needed. After the white paper has been accepted by program management (including the MEPAG executive committee), it will be posted on a publicly accessible web site.

The report should not include any material that is a concern for ITAR (as is true of everything done by MEPAG).

Note, the bulk of this work and the draft white paper was completed by September 2004. There have been unavoidable delays to its publication. In the meantime thinking about AFL has progressed. This document reflects the thinking in September 2004. Whilst engineering and programmatic changes have occurred since then, the strength of this document lies in the science definition for the mission.

# DEFINTIONS

During the course of the AFL-SSG discussions several questions related to the MSPSG statement arose. Specifically these questions relate to the definitions of, for example, the terms habitability (or what constitutes a habitat) and biosignature. Critical questioning by the group resulted in the formation of a definitions subgroup

The following definitions were decided upon by that group. These definitions are consistent through this document and although we cannot suggest the wider community adopt these definitions it is suggested that some consensus within the MEPAG members is reached to prevent numerous iterations of this process in other reports.

Abiotic Chemistry

Mainly carbon based chemistry the speciation and composition of which has remained simple with the production of all different isomeric possibilities and show no chiral or species preferences. In this scenario complex molecules may only be kerrogenous in nature (type iv) and similar to that found in meteorites.

Biosignature

Any phenomenon produced by life (either modern or ancient). Two sub-definitions: Definitive Biosignature: A phenomenon produced exclusively by life.  Due to its unique biogenic characteristics, a definitive biosignature can be interpreted without question as having been produced by life. Potential Biosignature: A phenomenon that may have been produced by life, but for which alternate abiotic origins may also be possible.

Extant life

General reference to living or recently dead organisms which may also possess a fossil record.

Extinct life

General reference to past life (and no longer present on the planet). If evidence remains, it is ONLY fossil.

Habitability

A general term referring to the potential of an environment (past or present) to support life of any kind. In the context of planetary exploration, two further concepts are important: Indigenous habitability is the potential of a planetary environment to support life that originated on that planet, and exogenous habitability is the potential of a planetary environment to support life that originated on another planet.

Habitat

An environment (defined in time and space) that is or was occupied by life.

Life detection

The process of investigating the presence of biosignatures (including potential biosignatures). Life detection can apply to either past or present life.

**Micro BioSensors (not to exclude organic chemical detection)**

Miniaturized instruments or instrument suites that are developed from technology such as Micro Electronic Machine Systems (MEMS), Micro electronic optic systems (MEOS), Microfluidics, Micro Total Analytical Systems (uTAS) or Lab-on-a-Chip (LOC).

Prebiotic Chemistry

Mainly carbon based chemistry the speciation and composition of which has a complexity and has produced a number of polymeric systems that could be used for structural, metabolic processes and information storage and retrieval.

Present life investigation

One that specifically targets living or recently dead organisms. Time resolved studies on seasonal and daily (with perhaps higher frequency) time scales may be required to confirm observations that a biosignature of present life has been detected.

Preservation Potential

The potential for a particular biosignature to survive and therefore be detected in a particular habitat.

Primary Sample

Geological material (e.g. rock, regolith, dust, atmosphere, ice) acquired from its natural setting on Mars.  Note: specific locations where data are collected by contact instruments are referred to as "targets", not samples.

Secondary Sample

Any sample derived from the primary, including splits, extracts, sub-samples, etc.

# 4.0INTRODUCTION

The primary science driver for the mission concept was to define the first Mars mission to concentrate fully on Astrobiology science goals (as defined within the recently updated Astrobiology roadmap). Therefore, to define the preliminary general science objectives, and the science floor, the level below which the mission is not worth flying. The Astrobiology Field Lab was created as a concept by the Mars Science Program Synthesis Group (MSPSG) during their Pathways planning discussions in 2002-03 and can be paraphrased as;

**Astrobiology Field Laboratory.** “This mission would land on and explore a site thought to be a habitat. Examples of such sites are an active or extinct hydrothermal deposit or a site confirmed by MSL to be of high astrobiological interest, such as a lake or marine deposits or a specific polar site. The investigations would be designed to explore the site and to search for evidence of past or present life. The mission will require a rover with “go to” capability to gather “fresh” samples for a variety of detailed *in situ* analyses appropriate to the site. *In situ* life detection would be required in many cases.” (*From MSPSG (2003)*

However, MSPSG deferred to a successor team (AFL-SSG) the definition of AFL’s specific scientific and engineering constraints, possibilities, and priorities. The AFLSSG team was initially convened in October 2003 and operated through a number of telecons and one face to face meeting. Therefore this team was asked to plan during a constantly shifting science focus and have constantly endeavored to keep abreast of the Mars Exploration Rover findings and review the goals and outcomes of the SSG accordingly. Undertaking this activity at a time when 3 new space craft have started to explore Mars has been exciting, inspiring and already produced new evidence to which we have responded. Many notions of how to perform this mission have therefore been updated from preconceived notions held before specifically, the MER data was returned. We hope that these changes reflect a renewed sense of optimism and realization of the location of interesting samples to interrogate with instrumentation currently under development.

# 5.0SCIENCE GOALS

## 5.1 Assumptions

To undertake this task the AFL-SSG was asked to consider the following assumptions;

Assume AFL will need to be ready to launch as early as the 2013 opportunity

Assume all missions scheduled before 2013 are successful.

The MSL entry-descent-landing (EDL) system has successfully been demonstrated, and the engineering heritage can be used on AFL.

Assume the primary goal of AFL is to make a major advance in astrobiology.

Assume a cost cap approximately equal to that of Ground Breaking Mars Sample Return.

These assumptions are based on the timeline suggested by the Pathways SSG, summarized in Figure 1.

Figure 1. A summary diagram of the pathways proposed by MSPSG.

Generated

From Figure 1 it can be seen that the pathways leading to AFL are propelled by the discoveries of hydrothermal habitats and the search for evidence of past life. During the course of the AFL-SSG discussions several questions related to the MSPSG statement arose. Specifically these questions relate to the definitions of, for example, the terms habitability (or what constitutes a habitat) and biosignature. Critical questioning by he group resulted in the formation of a definitions subgroup the results of which are shown in Section 2.

Responses to discoveries other than pathway to discover hydrothermal habitats as shown in Figure 1 were deemed necessary and led to the formation of the hydrothermal, ice, sedimentary and water subgroups. Through these discussions the parallel nature of exploration and engineering goals in different environments was explored and a “core” of similar themes and objectives arrived at that included life detection philosophy, measurements, rover capabilities and sample preparation. This notion is explored further in section 8.1.2.

Other questions arising from the MSPSG guidelines and our discussions related to “the capability to gather fresh samples” which led to the formation of the sample preparation subgroup. The mention of *in-situ* life detection led to the Instrument subgroup surveying and documenting the current instruments in development.

Several assertions for the completion of these science goals were formulated and are as follows:

1.By 2013 a full model of the potential habitability of Mars, organized by environment, and applicable to both the present and geological past will be partially understood. Therefore the Mars program will have to choose to either; select one environment with a high habitability potential and test for habitation *or* continue to refine the habitability models to allow better targeting of a subsequent habitation mission.

Therefore we forecast one of two conditions will be true in 2013:

•More likely: Models of habitability require either further definition or further confirmation before a specific test for habitation should be attempted.

•Less likely: At least one environment (past or present) with high habitability and preservation potential has been identified, and a habitation test is justified.

We therefore questioned whether AFL would be effective in both scenarios. Which further reinforced the concept of defining a core set of mission parameters (Section 8).

2.Organisms and their environment together constitute a system. Each produces an effect on the other. Some kinds of investigations can simultaneously provide information about both the environment (e.g. habitability potential) and associated life forms (habitation).

3.Traditional Mars mission planning has involved choosing scientific objectives and investigations for EITHER prebiotic chemistry, extinct OR extant life. (PP policy is structured the same way.) However, some kinds of scientific investigations will detect all of the above categories and potentially measure the signs of life without prior need to assume search parameters that will pre-categorize whether it is extant or extinct.

4.As our exploration of Mars (through robotic and sample return missions and terrestrial studies on Martian meteorites) proceeds, anomalous features will be discovered that are POSSIBLE biosignatures for Martian life forms. It is important that this

Observation of POSSIBLE biosignatures can be made by relatively simple observations (e.g. geological, textural, geochemical). Such features would constitute a working hypothesis, **NOT** confirmation that life exists and has been detected.

Concluding that evidence of a Martian life form (past or present) has been discovered requires proving that a POSSIBLE biosignature was produced by the activities or remains of Martian life. Unless a POSSIBLE biosignature is proven to be a DEFINITIVE biosignature – an object or phenomenon that could only have been produced by life – it may not possible to prove the presence or former presence of life on Mars using AFL alone. However, the AFL mission has been configured so that it will not miss POSSIBLE biosignatures if they occur in a similar habitat and with similar character to those found on Earth and may indeed detect those non-earth centric signatures that would, without prior knowledge of the state of an unknown biochemistry, appear to be reasonably measurable.

Once several POSSIBLE biosignatures are identified, additional efforts will need to be made to prove that they definitively represent extant life or former life, or determine whether the group of POSSIBLE biosignatures is CONSISTENT with the hypothesis that life exists or once existed on Mars.

The current MEPAG goals document highlights the following strategy for Goal 1 “The search for Life” Determining if life ever arose on Mars is a challenging goal. *The essence of this goal is to establish that life is or was present on Mars,* ***or*** *if life never was present to understand the reasons why Mars did not ever support its own biology. A comprehensive conclusion will necessitate understanding the planetary evolution of Mars and whether Mars is or could have been habitable and will need to be based in multi-disciplinary scientific exploration at scales ranging from planetary to microscopic. The strategy we have adopted to pursue this goal has two sequential aspects: Assess the habitability of Mars (which needs to be undertaken environment by environment), and in environments which can be shown to have high habitability potential, to test for prebiotic processes, past or present life. These constitute two high-level scientific objectives. A critical means to achieve both of these objectives is to characterize Martian carbon chemistry and carbon cycling. The science associated with carbon chemistry is so fundamental to the overall life goal that we have established it as a third primary science objective. To some degree, these overarching scientific objectives can be addressed simultaneously, as each requires basic knowledge of the distributions of water and carbon on Mars and an understanding of the processes that govern their interactions.*

Importantly this statement points out that the seemingly differing goals, habitability, Carbon chemistry and the search for biosignatures, overlap and can therefore be addressed to a significant degree by the interpretation of measurements undertaken by certain instruments. Examples, habitability demands the presence of Carbon, biosignatures are often Carbon based etc. Amino acid analysis, n alkane distributions, selection of informational and catalytic polymers based on a narrow range of particular molecules and isomers of a particular molecular group. For example nucleic acids contain ACTGU on earth, but may contain LMNOP on Mars, it is the presence of a narrow range of the possible purines and pyrimidines available through abiotic processes that would constitute a biosignature. This could be true of any potential novel biomolecule and it *may be* that upon detecting a small range of the possible isomers of a particular compound speculation as to their informational or catalytic roles can begin.

Therefore AFL can reasonably begin the process of life detection by characterizing potential biosignatures.

## 5.2 Objectives

Proposed overall scientific objective of AFL:

For at least one Martian environment of high habitability potential, further investigate the potential for habitability, the potential presence of the chemical precursors of life, the potential for preservation of biosignatures, and possible signs of life.

This objective must balance the need to be a significant extension beyond currently planned missions, yet not an unrealistic extension of current technology. The detailed objectives proposed include (in no order of importance);

1.Within the region of Martian surface operations, identify and classify Martian environments (past or present) with different habitability potential, and characterize their geologic context.

2.Quantitatively assess habitability potential.

Measure isotopic, chemical, mineralogical, and structural characteristics of samples, including the distribution and molecular complexity of carbon compounds.

Assess biologically available sources of energy, including chemical and thermal equilibria/disequilibria.

Determine the role of water (past or present) in the geological processes at the landing site

3.Investigate the factors that will affect the preservation of potential signs of life (past or present) on Mars

4.Investigate the possibility of prebiotic chemistry on Mars (including non-carbon chemistry)

5.Document any anomalous features that can be hypothesized as POSSIBLE Martian biosignatures. This will constitute a set of working hypotheses, which will need refinement and further testing on Mars or in return samples.

### 5.2.1 Habitability

A definition for habitability is contained in section 2. From the first assumption above the following recommendation was made:Habitability models have the potential to integrate many different classes of information that have been made recently and will be acquired over the next decade. However, they will be most effective if placed on a semi-quantitative footing (see Appendix II for an example).This question was then followed up in discussions within the definitions subgroup and illustrated by Figure 2.

Habitability should be described by measurable parameters that index the potential of an environment to support life. Only in this way can the scientific community achieve consensus regarding whether or not a given environment is habitable, either for Martian or Earthly life. For any living system, certainly there will be a range of environmental requirements, outside of which life will be unsupportable. Even though we have no information on potential Martian biological requirements, we can learn from universal Earthly life requirements. The AFL study group has agreed that Earth life requires water and certain chemical raw materials such as carbon, hydrogen, nitrogen, phosphorus and a few others in trace amounts (Williams and Fraústo da Silva, 1996). We also know that life makes products from these raw materials with the additional requirement for an energy source, so sufficient habitat space must be available for the products to be mobilized or diffuse away, otherwise metabolic reactions would run to equilibrium, or possibly reverse. On Earth, the chemistry of life involves oxidation-reduction reactions, and metabolism from the archaea to some highly-evolved eukarya requires electron donor/receptor pairs. The spatial distribution of both oxidized and reduced forms of ions involved in respiration may be as important as their concentration in the context of biological requirements.

We assume that the astrobiology community will have made progress toward consensus regarding the indexing of habitability before the launch of an AFL mission, as the concept of habitability will have an impact on missions with the scope of Terrestrial Planet Finder to SSE missions in search of present or past Martian habitable environments. One approach toward such progress may lie in development of terms that lead to a probabilistic evaluation—a scale of habitability based upon measurements of agreed-upon parameters such as threshold concentrations of water and other raw materials, energy, etc.

### 5.2.2 Extinct or Extant Life. Abiotic or Prebiotic Material

It is important to recall that life on Mars may be composed of many molecules that differ from those of Earth life. However, most current hypotheses on extraterrestrial life maintain that Martian life, if it exists or once existed, will resemble life on Earth in that it will be: 1) composed of carbon, 2) based on a ‘nucleic acid like’ replication mechanism and 3) packaged in cellular compartments. Measuring the distribution, isomerization and quantities of carbon species limits the search to life based on carbon chemistry, an appropriate goal that reflects the strategies used to locate the biosignatures of ancient carbon-based life forms on Earth. Potential organic carbon species that would need to be distinguished by AFL are given in Table 2.

In the search for biosignatures on Mars the interpretation of measurements will determine whether a particular results indicates the category to which a particular a/biosignature should be placed i.e. pre/abiotic extinct or extant. The important issue is to make the correct measurements to ensure the sensitive detection of molecules of interest can be undertaken.

Figure 2 Illustrated the cross cutting relationships between the searches for habitability in comparison with the search for evidence of past or present habitation.

From assertions 2 and 3 and illustrated by Figure 2 the implications are that***:***The distinctions between investigations of habitability potential, habitation, extinct life, and extant life are blurred. It is possible to configure a mission that has relevance to ALL of these subjects.

Without evidence of liquid water on Mars, the potential to locate extant Martian life is less, as all conceivable life forms require liquid water. Hence the focus of upcoming missions on determining whether liquid water is available. Until this information is known, an AFL mission will need to be prepared to detect both extinct and extant life, as well as be able to distinguish abiotic and prebiotic material. We assume that the investigation of abiotic and prebiotic chemistry will be useful in evaluating the postulated meteoritic and cometary delivery of exogenous organics to the lithosphere and the formation of organic material by indigenous hydrothermal processes. The current MER information that Mars harbors environments that contained liquid water in the past indicates that the possibility of discovering extinct life has increased.

All information gained from AFL will be useful with regard to either describing what kind of life exists/existed on Mars or describing conditions found on Mars and determine why life evolved on Earth and not Mars (assuming the conditions on Mars are similar to those on Earth). The search for the signatures of prebiotic chemicals or components of life–past or present will provide important information that will advance the field of astrobiology and the understanding of our own planet. In addition, there is now considerable evidence pointing to the presence of methane in the atmosphere on Mars (Kerr 2004a.b commentary). This implies that geological processes on Mars could provide a chemical potential and carbon source that could be used by microorganisms and may indicate the presence of hydrothermal sites and liquid water. The generation and fate of atmospheric methane on Mars would be a significant goal for missions that fly prior to AFL. Such measurements would significantly improve our understanding of habitability.

Investigating early planetary surface chemical processes on Mars is important to understanding two possible program-level exploration outcomes:

If life is not present at a specific test site, can we predict that it might exist elsewhere?

If life never formed on Mars, WHY?

Studying such issues will also address specific goals, issues:

Understand planetary evolution through elucidating organic chemical input i.e. meteoritic versus abiogenic synthesis reactions.

Mars may give clues to the prebiotic evolution of the Earth. On Earth an unaltered geologic record of early planetary evolution (4.5-3.8 Ga) does not exist.

Allow conjecture as to why life did not start on Mars (should that be the outcome). Were the chemical processes and building blocks present there as on Earth?

By definition, a biosignature is an indicator of life or biological activity. Therefore, by definition, the discovery of even one biosignature on Mars would indicate that life once existed on the red planet. However, discoveries of ancient POSSIBLE biosignatures on Earth and Mars have shown that it can be extremely difficult, if not impossible, to prove their biogenic origin. Our inability to prove an object or phenomenon’s biogenic origin (i.e., biogenicity) is hampered by the fact that inorganic processes can produce abiotic mimics of biosignatures. Hence the need to make a distinction between a POSSIBLE biosignature and a DEFINITIVE biosignature.

A DEFINITIVE biosignature is one that has attributes that can ONLY be produced by life or biological activity. Until such time that a POSSIBLE biosignature is proven to be a DEFINITIVE biosignature, the former constitutes a working hypothesis that requires additional characterization. AFL will contain the necessary equipment to detect POSSIBLE biosignatures (e.g., microfossils, biofabrics, biominerals, biomarkers, biomolecules isotopes, etc.). However, short of locating a living or perfectly preserved cell that displays the structural complexity indicative of biosynthesis, establishing that a POSSIBLE biosignature is DEFINITIVE evidence for life will require further testing. It will also be necessary to prove that a biosignature is indigenous to Mars and not a contaminant, regardless of whether we discover it on Mars or in rocks or sediment returned to Earth from a future sample return mission from Mars. These considerations underscore the need to distinguish a DEFINITIVE biosignature from a POSSIBLE biosignature. This underscored the goal of the definitions sub group that postulated that only by producing several mutually supporting lines of evidence (i.e. possible biosignatures) could a definitive biosignatures be postulated.

The lack of a conclusive set of criteria for life detection and preservation has been illustrated recently by two debates; the search for the oldest evidence of life on Earth and the raging debate on the claims for life in ALH84001 (McKay, 1996). The scientific controversies over the former debate, that of the earliest evidence of life on Earth, have recently intensified but are still unresolved (Schidlowski, 1988; Schopf, 1993; Mojzsis et al., 1996; Rosing, 1999; Mojzsis and Harrison, 2000; Brasier et al., 2002; Fedo and Whitehouse, 2002, Pasteris and Wopenka, 2003, Furness 2004). The common denominator in both of these debates is the underlying difficulty, or inability to demonstrate conclusively the biological origin of the respective evidence, which in either of the above cases would have to be seen as conclusively proving the presences of fossil microbial life. However, a consensus that has emerged from these discussions, and is now seen as a critical requirement, is the demand for further lines of evidence in addition to any morphological data that supports such extraordinary claims. Since the inception of the second debate, that of life in Martian meteorite ALH84001, it has become evident that there is no consensus on the nature of life in extraterrestrial materials. Indeed techniques supposed to detect life failed, for whatever reason, to conclusively detect the presence of terrestrial organisms within this meteorite (Steele et al., 1999, 2000, Toporski, 2000). Recent studies suggest that the mass spectrometry experiments on the Viking lander would have missed 3x107 bacteria per gram of Martian regolith (Glavin et al., 2001). These examples are beginning to show that only by means of a multi-disciplinary, multi-instrument scientific approach, will the above questions be answered. It is clear that a great deal of additional systematic experimentation and testing must be undertaken in terrestrial environments to better determine the criteria by which biogenicity and therefore preserved biosignatures can be quantified.

Though there are a number of ways of categorizing biosignatures, microbial biosignatures found in ancient Earth rocks can be organized into three categories: *bona fide* microfossils, microbially influenced structures, and chemical fossils, also known as chemofossils (Cady et al., 2003). Bona fide microfossils, which may include cellular and/or extracellular remains (e.g., carbonaceous microfossils), display structural and chemical characteristics that confirm their biological origin. Microbially influenced sedimentary structures (e.g., biogenic stromatolites and microbialites), display biofabrics and morphologies known to have been produced by the presence and/or activity of biofilms or microbial mats. Chemofossils (e.g., biomarkers and biominerals), display chemical, isotopic, and structural characteristics indicative of biological activity.

Among the chemical biosignatures that have been identified as applicable to past and present biological activity on Earth are the biominerals, that is, minerals formed by biotic processes, either directly, or indirectly. Biominerals have been found in the fossil record that date back to the Precambrian. It has been suggested that biominerals could be important indicators of life and thus could play an important role in the search for past or present life on Mars (Schwartz et al., 1992, Cady et al 2003). Furthermore, organic components (biomarkers) that are often associated with biominerals are believed to play crucial roles in both pre-biotic and biotic reactions. For measurements carried out on Mars, a crucial step will be the in situ quantification of the nature, structure and concentration of biosignatures as a function of depth and time.

|  |  |
| --- | --- |
| **Source of Carbon** | **Carbon compounds. examples/comments** |
| Abiotic molecules from meteoritic / cometary influx | Amino acids, purines and pyrimidines, polycyclic aromatic hydrocarbons, chain hydrocarbons, fatty acids, sugars and sugar derivatives. |
| Prebiotic/abiotic molecules from synthesis reaction process on Mars | Amino acids, purines and pyrimidines, polycyclic aromatic hydrocarbons, chain hydrocarbons, fatty acids, sugars and sugar derivatives. |
| Terrestrial contaminating organics | Condensation products derived from rocket exhaust, lubricants, plasticizers, atmospheric contaminants |
| Terrestrial contaminating organisms | Whole cells, cell components (LPS, DNA, proteins, cytochromes) found on AFL itself. |
| Terrestrial like organisms – from Earth | Organisms not present on the craft measuring them, but had been previously transferred from Earth by either meteorite impact or contamination of previous spacecraft. Target molecules could include individual genes, membrane constituents, specific enzymes, and co-enzymes that would be expected to be over expressed or adapted in Martian conditions |
| Terrestrial-like organisms – evolved on Mars | Organisms that utilize terrestrial like biochemistries and have evolved on Mars Target molecules could include individual genes, membrane constituents, specific enzymes, and co-enzymes that would be expected to be over expressed or adapted in Martian conditions or organisms using metabolisms that would not be present on a space craft contaminant such as methanogens, psychrophiles endolithic survival mechanisms. |
| Non-terrestrial-like organisms | Utilizes an array of molecules for information storage, information transfer, compartmentalization and enzymatic activity that differ from those used by extant terrestrial life. Examples would be the use of novel amino acids and nucleotides or the use of novel nitrogen utilization strategies. |
| Fossil biomarkers | Detection of established terrestrial fossil biomarkers such as hopanes, archaeal lipids and steranes, for the detection of the diagenetic remains of terrestrial based life. Characterization of potential breakdown products that can be reasonably extrapolated from the detection of molecules comprising an extant Martian life form. Detection of the diagenesis products of extinct Martian organism based on carbon compositions consistent with biological fractionation of a narrow range of abiotic precursors. |

|  |  |  |  |
| --- | --- | --- | --- |
| **Component** | **% Total Weight (or mass C x10-13 g)** | **Number of Types** | **Preservation** |
| **Water** | 70 (NA) | 1 | Unknown in Organic and mineral phases |
| **Proteins** | 15 | ~2000 | 1000’s without protection by a mineral matrix. ~45Ma with protection?. |
| **Nucleic Acids** |  |  |  |
| DNA | 1 | 2+ | Oldest ? ~350,000 |
| RNA | 6 | (see below) | Days – Months (studies on longevity of RNA other than in clinical settings have not been performed. |
| *rRNA* | 5.5 | 3 | Days – Months |
| *tRNA* | 0.1 | ~32 | Days – Months |
| *mRNA* | 0.3 | 1000’s | Days – Months |
| *Non coding RNA* | 0.1 | 1000’s | Days – Months |
| **Polysaccharides** | ~1 | Uncounted | Chitin - 25Ma. Exopolymer sheaths ~2Ga |
| **Lipids** | 2 | ~50 | Cell wall components - Hopanes 2.7Ga |
| **Amino acids** | 0.4 | ~100 (20 main ones) | As protein diagenesis – Ma.  Chiral signal in fossils lost after ~ 1 Ma. |
| **Sugars** | ~3 | ~200 | Days to weeks (see polysaccharides) |
| **Other small organics** | 0.2 | ~200 | Porphyrins ~ 2 Ga |
| **Inorganic species (C, H, N, O, Fe, P, S etc).** | 1 (~100% dry weight) | ~20 – 30 (including inorganic complexes) | Isotopes may preserved for ? 3.5 Ga for C.  Research is continuing to define other isotope systematics for preservation of a biogenic signature. |
| **Diagenetic Macromolecular material** | Total cell breakdown products (100% dry weight of cells) | Kerrogens (4 types)  Melanoidins (100’s) | Kerrogens – ? 3.5Ga for biogenic (Type 1-3). Type 4 indicative of meteoritic input.  Melanoidins conbination of sugar and proteins, ~50 Ma. |

Generated



t.

One underling theme of astrobiology is the differences in planetary evolution and how that relates to habitability of planets. If Venus, Earth and Mars all formed in the “Habitable zone” of the sun why is Earth the only one to be teaming with life? An AFL mission to high northern latitudes can help elucidate this concept, by helping to understand both geologic and climate changes on Mars over it’s history.

Ice exists on Mars in vastly different geologic settings and therefore there are several major differences in the science requirements both with respect to ice bearing regions as well as other Martian regions (i.e. sedimentary and hydrothermal environments). Here we will discuss science requirements that span the different geologic settings, above and beyond what the core AFL science requirements. As mentioned previously life can exist in these locations by either becoming dormant until conditions exist where the temperature is above freezing point of water, or by creating pockets of liquid water by lowering the freezing point of water. Determining if an acquired sample contains liquid water requires the collection of sample without raising the temperature above the local melting point of water (keeping in mind that the concentration of brines in the sample can dramatically lower the melting point below 0°C). The determination of liquid water in a sample is not necessarily a measurement of life, because liquid water can exist in meta stable state in some Martian environments without being associated with life (Hecht 2002). However, samples containing liquid water would be a priority target to be analyzed by the analytical laboratory instruments. In the Northern polar layered deposits the measurement of strata of layered terrain to see potential differences in layering and effects due to Aeolian processes. This would require imaging at several spatial scales.

A determination of the yearly cycling of CO2 and H2O will not only lead to a better understanding both current and past atmospheric dynamics (Clifford, Crisp et al. 2000) it can potentially identify if a biosphere is in contact with the surface elsewhere on the surface. Recent discovery of methane in the atmosphere from both ground based observations and from the Planetary Fourier Spectrometer (PFS) onboard the ESA’s Mars Express, although most likely not from biologic process, demonstrates that a better understanding of atmospheric process are needed (Kerr 2004a,b). If biology is in contact with the atmosphere, this maybe detectable from orbit (i.e the recent measurements of methane) but whether life produced these gases can only be ascertained by painstaking surface measurements.

*Science Trades*

Because potential ice missions have different geologic regions there are several science trades that can be made so as to maximize the science return of the mission. The first science trade that can be made is the level of mobility requirement. For missions to the permafrost regions and on the polar caps potentially require very little mobility (only 10’s to 100’s of meters) depending on high resolution orbital mapping by Mars express and Mars Reconnaissance Orbiter. Current orbital data on those scales indicate not much difference in geologic setting over km distances. Therefore large surface mobility could be not as scientifically important as it is for other regions. There would be, however, a need for greater subsurface access including drilling well below 1 meter to increased ice concentrations. Therefore a potential trade of horizontal distances vs. depth, would need to be made

On the other end of mobility spectra is the recent ground water site which can require large “goto” capability of at least the level of the landing precessions if of a landing ellipse can be placed near that site. This may require mobility in the 10’s of km, similar to what would be required in the sedimentary region.

The nature of high latitude northern sites indicate that for extended missions nuclear power is most likely the only feasible alternative for mission power generation as Mars progresses through its year. However, for more equatorial missions solar power can be a feasible alternative especially given the projected longer lifetimes that the on going MER missions are demonstrating. This trade will depend on the expected duration of the mission and ground operations and ability to land at high latitudes as set forth in the science requirements.

*Site Specific Measurements and sample handing and preparation requirements*

Measurement requirements are dependent on location. The measurements that are required for ice missions resemble the instrument complement for the other missions scenarios postulated (hydrothermal, and sedimentary deposits) and the measurements requirements can be found in section 8.2. Here we discuss measurement requirements specific to ice regions.

Remote instruments

Mast based instruments must be able to do visual site reconnaissance at a level at least as well as PanCam on MER. Identifying potential targets from the distance of a daily traverse should be a requirement so that interesting samples can be targeted. Remote mineralogy of potential samples from a distance of 10 meters so that samples can be identified. The remote mineralogy instruments may have to account for ground frost when choosing a spectral range for a mast-based instrument. These requirements are virtually the same regardless of the environment AFL explores. In addition, if AFL is going to perform subsurface sample acquisitions in a high H2O environment, some subsurface reconnaissance must be done, especially if H2O varies dramatically in depth over 1 meter scales. A body-mounted detector capable of reconnaissance styled elemental abundances would also be desirable measurement if feasible and kept within the cost cap of the mission. This measurement could detect high potential astrobiological sites, as well as ground truthing orbital data. Finally, for polar cap missions, the cycling of H2O and CO2 and the interaction of those molecules from the surface to the atmosphere needs to be determined. The Martian atmospheric dynamics is not currently in equilibrium (Clifford, Crisp et al. 2000) (i.e. Aeolian processes, ablation and sublimation) Determining the atmospheric polar properties can help put a constraints on atmosphere compositions and help determine if a biosphere presently exists, as well as long term possibilities that a more favorable climate once existed. This is especially true given the recent detection of methane in the atmosphere at trace levels by both ground and orbital observations.

Contact Instrumentation:

The instrument delta between AFL ice and other AFL missions is that direct detection of liquid water present in a sample needs to be made. The Phoenix lander is attempting to make this measurement as well, and lesions learn in that mission will affect the design of this measurement. For mission to the polar cap, any contact instrument will also have to account for the ice core that is being obtained.

Sample Acquisition and Processing:

All of the hardware infrastructure referred to in this environment must be able to handle relatively large amounts of water. This includes the drills, corers, and precession sample processing and distributions stations. Water can interfere with the drilling process either by making material harder to drill into or by melting and creating a mud like material that can interfere with machinery. Drilling into this material without melting the water or using drilling fluids will need to be developed and demonstrated in both a relevant terrestrial environment and under simulated Martian conditions. Finally, for missions to the polar cap, a different sample acquisition system will need to be developed. This instrument will have to be able to melt and sublimate any CO2 or H2O while collecting impurities in the ice material.

## 7.4 Water

*Science Theme*

Assess present (*and past?*) Martian astrobiology by studying liquid water in the shallow subsurface.

*Proposed science strategies*

Drill, core, or otherwise obtain liquid water sample.

Determine concentrations of redox sensitive aqueous compounds, including O2, H2, HCO3-, NO3-, Fe2+, SO42-, H2S, NH4+.

Determine presence (if possible, concentrations) of DOC and aqueous organic monomers, including carboxylic acids, amino acids, sugars, hydrocarbons (or should be target functional groups instead?).

Determine presence (if possible, sequence or composition) of organic polymers, including proteins, lipids, nucleic acids.

Visualize microbial cells (if present) with light microscopy on stained and/or unstained cells.

Carry out microculturing on 1-3 samples using tens to hundreds of pre-designed growth media at several different temperatures.

# 8.0Core Mission Components

As discussed in sections 6 and 7, there currently are multiple possible variations on the AFL mission theme. Opinions differ as to the specifics of these variations in terms of context and priority, which may lead to revisiting the chosen site if selected. However, the AFL-SSG feels that it is possible to define an invariant core, which is common to most versions, along with a discovery-responsive and competition-responsive cap.

The proposed mission requirements to ensure the greatest scientific return for the AFL mission include:

“Go-to” mobility (ability to access a specific target).

When sites are identified from orbit that possess high astrobiological interest (see Section 6.0) the rover has to be able to access them, even if the nearest safe landing site is 10’s of km away. The rover also has to explore several different regions within a high interest site. An example of this is Holden Crater (see Section 7.1) in which what resembles an ancient river delta is clearly visible in orbital images. Exploring the specific features found there would require not only a landing ellipse directly outside the feature but the ability explore several different locations several km’s apart within the potential delta system.

+60 to –60 (seasonal polar cap) for sedimentary/hydrothermal. +45 to +85 for ice mission (See section 7.3).

Precision landing (1 km) and the ability to land in terrain that is rougher than we have targeted in the past (hazard tolerance, hazard avoidance).

In order to access more of the planet for exploration by AFL, as well as limiting costly “Go-To” traverse, having a suitable landing ellipse smaller then 10km is required. This enables access to regions like Melas Chasma, where suitable landing ellipses greater then ~5 km prove difficult to identify.

Subsurface access of 1-3 m, and multiple holes. Probably also have a need to expose / drill into material in outcrops .

Organic material on the Martian surface may be extremely scarce, primarily due to an oxidizing layer thought to exist because of UV fluxes at the surface. How far down this oxidant penetrates is not presently known or constrained, therefore shallow (<3 meters) subsurface material may be void of organic material. Accessing and analyzing this material may indicate if extant life is possible in a protected subsurface environment. However, if the surface regolith is largely made-up of unconsolidated material, organic free material may be thoroughly mixed by several billion years of global dust storms. In this scenario all organic material may have destroyed down to >3 meters, making analysis of this material a lower propriety (hence not a requirement). Subsurface access of potential bedrock and out-crops is highly desirable in any scenario where it is present.

Organic contamination: be able to collect and deliver Earth-clean samples to on-board laboratory

It is a requirement to have samples that are not contaminated by terrestrial organics to a level greater then the minimum level of detection of the astrobiology specific instruments. See report of the Organic contamination Science Steering group (Mahaffy et al., 2004).

Sample preparation including spatially controlled precision sub-sampling and liquid extractions for selected high-potential samples.

The AFL-SSG has determined that identifying the best possible sample for analysis is a primary requirement for a future AFL mission. See section 8.3 for a discussion of these requirements in more detail.

## 8.1 Payload strategy

It was determined that payload characteristics could be defined as core to any potential AFL mission concept as described in Section 7. These include:

Acquiring the right sample.

In order to maximize the probability of detecting biosignatures in a ***location with the high general habitability potential*** has to be identified. Several of the reconnaissance missions (see section 6), will be used to identify this location. In identifying the location, the ***understanding of the preservation potential*** of this location must be better understood. The Earth is inundated with biological material, where most (if not all) sites on the surface (and possibly the subsurface) should have a continual influx of biologic material. On Mars this is not the case. A location on Mars which once supported life, may not have any record of that life, due to chemical interactions, or by meteoritic impacts. Understanding how a site on Mars preserves a record of past life is essential toward acquiring the right sample. In this regard there is the need to be able to access samples with the highest probably of being astrobiologically important. This includes both ***identification of specific samples*** as well as the ***ability to acquire that sample***.

Understanding the geological, mineralogical, and chemical context of that sample

The labeled release experiment aboard Viking, released nutrients into a Martian regolith sample to determine if metabolism took place. The results of this experiment on their own can indicate that metabolism was taking place. However when taken with the GC/MS data it was generally understood that a chemical reaction was taking place within that sample due to the oxidants present in the surface material (Mancinelli 1998). A complete understanding of the relationships between geological, mineralogical and chemical characteristics of the sample is needed to determine Astrobiologically implications of analytical measurements.

Identifying the best place on the sample

Instead of introducing a core into a bulk rock crusher, in which most of the material will not be analyzed, it was determined that sampling of small features of a sample would be required. Section 8.3 describes this precision sub-sampling in more detail.

Performing at least 3 different Astrobiologically related measurements.

The detection of biosignatures on Mars would, to put it mildly, fundamentally change our perception of life else where in the universe. In order to avoid ***potential false positives***, three separate measurements would need to be preformed on a sample to confirm any one measurement. Furthermore, repeat measurements will also help to avoid ***false negatives***. Since Martian life may be very different from terrestrial life, different measurement techniques may return a positive, while others measurements may miss more subtle signs that life is present in the samples. If one or two instruments detect interesting signatures, future missions can be designed to further explore the same site for these signatures.

## 8.2 Core Measurements and Instrumentation

As stated in Section 5.2, the proposed overall scientific objective of AFL is, for at least one Martian environment of high habitability potential, to further investigate the potential for habitability, the potential presence of the chemical precursors of life, the potential for preservation of biosignatures, and possible signs of life. This is to be accomplished through measurements supporting the following (un-prioritized) detailed ***Mission Objectives***:

Within the region of Martian surface operations, identify and classify environments (past or present) with different habitability potential, and characterize their geologic context.

Quantitatively assess habitability potential:

Measure isotopic, chemical, mineralogical, and structural characteristics of samples, including the distribution and molecular complexity of carbon compounds.

Assess biologically available sources of energy, including chemical and thermal equilibria/disequilibria.

Determine the role of water (past or present) in the geological processes at the landing site.

Investigate the factors that will affect the preservation of potential signs of life (past or present) on Mars.

Investigate the possibility of prebiotic chemistry on Mars (including non-carbon chemistry).

Document any anomalous features that can be hypothesized as possible uniquely Martian biosignatures. This will constitute a set of working hypotheses, which will need refinement and further testing on Mars.

The following ***Measurement Requirements*** for the AFL Core, derived from these objectives, were specified in order to support the instrument development and selection process for AFL:

**Comprehensive Imaging** - Fully image the overall landscape and each investigation scene to assess the variety of local environments (past or present) that can be discerned from expressed surface features such as outcrops. Include both color optical stereo imaging and higher-resolution long-focal-length telescopic imaging of key areas of high interest for further investigation of habitability potential. Target range is 1 m to infinity/horizon. High magnification or high resolution imaging should be able to discern layering at the 10 cm scale from a distance of 1 km. These measurements support the decision to focus more closely on specific sites, targets, and samples. *Supports Objectives: 1*

**Definitive Mineralogy and Chemistry** - Determine mineralogical and chemical (elemental) composition at all scales of investigation: *site/scene surface reconnaissance scale* (range: infinity/horizon to meter; resolution: km to cm), *hand-sample scale* (range: meter to cm; resolution: cm to mm), and *acquired subsample scale* (bulk measurement of a few-mm subsample with high accuracy), with respectively increasing degrees of definitiveness and sensitivity. *Supports Objectives: 1, 2, 3.*

**Redox Potential** - Assess the redox potential and oxidation chemistry of the near-surface environment. This measurement details how much energy is available for an organism to use in growth and reproduction and would be required to be measured to a precession of 10 mV. *Supports Objectives: 2, 3*

**Fine-Scale Surface Analyses** - Investigate the surfaces of exposed or acquired samples at fine scales for morphological, chemical, and molecular signatures suggesting preservation of pre-biotic or biotic organic compounds. This may include directly-detected compositional markers, evidence of minerals formed in or altered by liquid water, or particular sample textures (i.e. concretions). Color optical imaging with resolution below 30 m (although for bacterial analysis in anything other than a macroscopic biofilm structure this would be inadequate) within a larger field of view should provide the context for co-focused spectroscopic tools such as UV-excitation fluorescence, laser Raman, or other fine-scale techniques to perform chemical signature detection. Spectroscopic tools must be able to analyze mm-scale regions on surface or drill core samples (e.g., through a focused excitation source or through high imaging/detector resolution). These surface measurements provide first-order astrobiological analyses and support the intelligent selection of subsamples to be processed in the analytical laboratory. *Supports Objectives: 2, 4, 5*

**Subsample Biosignature Analyses** - On selected subsamples, perform an array of high-sensitivity, mutually-confirming laboratory investigations related to astrobiology goals. *Supports Objectives: 4, 5*

The identity, abundance, and isomeric distribution of carbon compounds should be thoroughly analyzed to low detection levels (ppb or below by weight within bulk ~102 mg subsamples) and to high molecular weights (hundreds to thousands of Da) at high peak resolutions (~2000 FWHM). Measurements utilizing broadband techniques such as pyrolysis GC-MS should be configured to enable the detection of less volatile species that are particularly relevant to determining preservation of biosignatures.

The isotopic ratios of H, C, N, O, and S should be characterized with sufficient precision to enable biogenic, environmental, or meteoritic fractionation trends to be identified based on requirements determined from MSL and other measurements (sub-per-mil to % levels). Compound-specific 13C/12C ratios coupled to the analyses in (1) are highly desired. Additional isotope ratios that further characterize atmospheric loss and other environmental fractionation processes relevant to astrobiology are also desired. Analyses may also be conducted on atmospheric samples to provide a more complete understanding.

Highly sensitive tests for the presence and characteristics of specific biosignatures should be conducted on bulk subsamples or isolated downstream extraction products (e.g., phases or concentrates). Biosignatures of particular interest include molecular compounds (or abundance patterns thereof) of distinctly biological origin as known on Earth, indicators of extant metabolic processes such as disequilibrium chemistry (molecular, biogeochemical, agent response, etc.), as well as chemical and morphological traces of such compounds and processes as preserved in the mineralogical microenvironment sampled. While the specific tests to be conducted will depend on the chosen AFL landing site and previous mission results, *examples* include detection of amino and nucleic acids, lipids, and proteins (with ppt detection limits if possible); chirality of amino acids and sugars (with %-level enantiomeric excess detection sensitivity); detection of concentrations of distinct molecules or isomers of the potential abiotic inventory that may represent the use and or concentration of a fraction of the molecules available through non biological interactions and finally direct detection of microbes, cells, or their fossils.

It must be mentioned that the advent of micromachining and the concept of micrototal analysis systems (uTAS) mean that through miniaturization the payload described may be integrated into a very small space whilst retaining accuracy and possible increasing analysis times.

The above information is summarized graphically in Figure 5.

Within the proposed AFL strategy, techniques to address the above requirements are structured in “tiers” following the expected level of physical sample contact: remote/standoff; contact; and laboratory. In the remote/standoff tier, the target “sample” is a wider area and not acquired by definition. In the laboratory tier, a small sample of interest has been acquired and possibly subjected to a preliminary analysis that supported the decision to subsample and deliver it to the laboratory for further analysis. However, in the contact tier, the sample may be analyzed before or after it is acquired (or both). This is designed to allow multiple levels of “triage” for determining the appropriate course of action with a given sample. An example of a post-acquisition contact measurement is a point-by-point imaging and chemical analysis along the surface of a several-cm long core. Based on this analysis, it may be decided to grind and/or otherwise process some or all of this core for analysis in the laboratory. For a description of the suggested mapping of measurements onto instruments placed in each of these tiers, refer to Section 8.1.4.

For completeness, the connection between the *AFL measurement strategy* and the *mission objectives* may also be characterized by indicating those objectives addressed while conducting the following activities:

Acquire the right samples (primarily 1; also 3)

Understand the context (primarily 1, 2; also 3, 4)

Identify the best place on the sample (primarily 5; also 2-4)

Perform mutually confirming astrobiology measurements (primarily 5; also 2-4)

This is summarized in Figure 5.

As mentioned above the instrumentation recommended for the Astrobiology Field Laboratory is divided into three categories or tiers: 1) remote sensing instrumentation located on a deployed mast, 2) a contact instrument suite located on a robotic arm, and 3) the laboratory suite located inside the rover and/or platform and fed with a sample acquisition and distribution system. The remote sensing suite is used to provide site characterization and rover navigation targeting. The contact suite performs “triage” analyses, mimicking a field biologist/geologist. The laboratory suite performs the detailed biology, chemistry, and mineralogy experiments required to quantitatively assess samples for past or present biological potential. Sample analysis instruments are supported by sample acquisition and processing infrastructure such as an articulated corer, (cm to 1 m) a rock abrasion/polishing tool, a precision subsampling tool, and possibly a 2.5 m drill.

The remote sensing suite includes at a minimum a panoramic multi-filter camera system that is used for site characterization, rover navigation, and first-order target selection. Additional instrumentation that may also be desirable may include reconnaissance-scale chemical and mineralogical experiments, such as hyperspectral imaging, stand-off (multi-meter) laser induced breakdown spectroscopy with fluorescence and Raman detection, and thermal infrared mapping for identifying geothermal sources of heat within the near-horizon of the Martian environment.

The contact suite must provide the second order triage for sample selection. The analogy is the selection and preliminary analysis of a surface material or hand sample by a field biologist or geologist. A sample arm equipped with an articulated coring drill and a rotating abrasive tool for clearing and polishing rock surfaces is envisioned for contact arm infrastructure. The contact suite includes at a minimum a course resolution (~20 m) microscope to examine the texture and other features of rocks and fines. Sample triage on AFL will however require additional contact instrumentation that further identifies materials of high interest for subsequent precision subsampling and laboratory measurements. The complement of contact instruments will be determined by the objectives at the type of site chosen for AFL: sedimentary, hydrothermal, ice, or liquid water. Possible arm-mounted spectrometers include: near infrared reflectance, Raman, Mössbauer, APX, deep-ultraviolet fluorescence, and/or various types of laser ablation sampling spectrophotometers and direct-inlet mass spectrometers. These tools are used to probe for and characterize samples of potential biological interest that may be delivered to the laboratory analysis portion of the payload.

Figure 5. AFL Measurement Requirements

Generated

The presence and design of the laboratory portion of the AFL payload is predicated upon a high degree of flexibility with respect to sub-sampling of the acquired rock core or soil sample. Therefore, there should be a strong emphasis on an *integrated* analytical laboratory approach to fully characterize common or related sub-samples: using microscopy as the “eyes”; definitive mineralogical and chemical identification from techniques such as x-ray diffraction, x-ray fluorescence, and laser ablation; and organic chemical and stable isotopic analyses that include at a minimum instrumentation capable of similar measurements to a pyrolysis-gas chromatography-mass spectrometer. Enhanced capabilities for identification of trace pre-biotic or biochemical compounds may be provided by staining followed by fluorescence detection techniques, solvent extraction/derivatization followed by a suitable ion mobility or mass spectrometry system, and other more specific techniques that target the detection of biomarkers such as amino acids, proteins, and/or DNA such as capillary electrophoresis, use of specific probes i.e. polymer or antibody systems and chemical assays. The particular implementation of more-specific biological/chemical analyses will depend both on the results of prior missions, such that their design and interpretation is advised by a solid first-order organic chemical characterization of Martian surface samples, as well as through analog field experiments targeted at terrestrial extremophiles. Additional capabilities such as detection of enantiomeric excess (chirality), rock dating, and fine-scale chemical imaging would be strongly complementary to the laboratory suite and highly desired for AFL. Such experiments might be provided by enhancements of previously mentioned instruments or by additional instruments.

The final selection of instrumentation on AFL will be based on a careful cross-matching of measurement requirements to instrument capabilities. It is recommended that the payload resources (mass, power, cost), and thereby the mission scope, for AFL be fundamentally and primarily driven by the sample preparation and instrument needs that are required to fulfill the measurement goals, rather than vise-versa. New instrumentation techniques as well as methods to optimally integrate techniques are desired and encouraged, but these must be maintained within a reasonable cost-risk profile. This necessitates a well-funded, well-advanced instrument development and integration program with strategic oversight form cognizant AFL program members.

The core measurements of AFL has been decided upon to answer the specific questions posed in the science rationale. The high number of instruments on this mission definition is a direct response to the findings of both sedimentary and hydrothermal deposits by the Mars Exploration Rovers and the subsequent realization that samples of Astrobiological interest may be much more accessible than originally thought. This allows deep drilling to be traded off against increased number of instruments.

## 8.3 Sampling and Precision Sub sampling

According to the various mission scenarios, different types of samples will need to be obtained, i.e. from rock, ice, regolith and sedimentary samples. The design of the SHAP facility and the exact number of samples to be handled and processed will depend on which mission scenario is decided on. This number will help define the sample collection system that will have to be developed. The basic concepts and design of the facility, however, will in principle remain the same for each type of sample and each type of measurement to be performed. Four different facets of the overall process are identified:

Obtaining a sample

Precision sampling of that core

Liquid and Heat extraction of organics

Contamination concerns

Issues were discussed with respect to each of these types of environments and are discussed in more detail in the following.

### 8.3.1 Obtaining a sample

Several sample acquisition tools are suggested for integration into SHAP facility. In order to be more precise the following defining terms have been made:

Corer: A device that can obtain a core which is ~ 5 cm in length with ~ 1 cm in diameter from an outer region of a rock on the surface of Mars.

Drill: A device, which can obtain sample from inside of a rock permafrost or sediment (cm – 1m) or from a distance underneath regolith (1-3 m).

Precision sub-sampling mechanism*:* A device, which can obtain a representative sample form a larger core. This would replace the rock crusher, which crushes the entire core, and produces fines, which might not be representative of the entire core.

Scoop: A device which can collect either fragments from a RAT, or unconsolidated regolith or permafrost material from the surface.

An aspect to consider is that idly sitting and drilling for significant time spans on Mars, drilling does not seem to be an efficient use of a limit mission lifetime. However, a system that could drill while not shaking the rover or consuming major power would need to be developed. For example, if an instrument such as XRD or deep UV fluorescence can analyze rock fines and fragments during drilling, we can identify samples that might be interesting, either because they are a fundamentally different type of mineralogy, or because of interesting organic components detected in them. This will not only help in sample identification, but also in the operations profile so that overall more samples can be analyzed.

Ice:

A drill would be required capable of gathering an ice rich sample while avoiding sublimating the ice, melting it, or volatilizing any constituent molecules. The sample must be as chemically similar to the material it is collected from in order to do a proper analysis. This most likely requires nighttime drilling operations in ice rich environments with a drill cooled to a temperature lower than that of the ice to mitigate cutting, melting and drill trapping problems .

Permafrost regions:

Samples that contain a mixture of ice and other material will have to be specially treated. Terrestrial permafrost can create problems when attempting to obtain a core from it in terms of both hardness and ability to keep the sample pristine. Samples will need to be obtained from a device that can be used multiple times while not heating the sample above freezing.

Surface Rock:

The depth MSL is coring to, 5 cm, seems to be a good number for the depth inside a surface rock that should be sampled. Terrestrial organisms practicing photosynthesis inside rocks inhabit approximately the outer 2 cm of sample a rock. Sedimentary material can be identified by obtaining several cm in length cores. It would be desirable to analyze the fines from the core if coring is progressing by a pneumatic device for introducing fines into the analytical suite. In this scenario, the XRD can analyze material as it is chilled in order to identify regions of interest, since the XRD can do infinite number of samples. When an interesting sample is identified it can be further processed via the other analytical instrumentation.

Regolith:

Regolith material would be obtained by a scoop attached to the rover arm and be delivered to sample processing facility. It is worth considering that if the regolith material is unconsolidated, we may assume that it was never in contact with the atmosphere. If it was at some point in the recent past, it could mean that it would be chemically the same as the surface material, and hence not worthy of a drilling effort. Unless the rover is going to head to a site where larger amounts of near surface (to the depth of the drill) water drilling into regolith would be desirable, otherwise it most likely is not.

### 8.3.2 Sedimentary deposits:

If a landing site were selected where drilling into for sedimentary deposits is required, it would be preferable to look at the entire core length. If a complete core could not be obtained because of engineering constraints, having a borehole instruments that can be lowered into the borehole, and examining the entire length of it would be very desirable. Instruments that could be utilized to answer astrobiologically relevant questions (rather then pure geologic questions) would need to be identified. In this scenario it might be worthwhile considering a drill/corer combination, where a drill takes 5 cm cores at a time, delivers to the sample processing facility and be lowered back into the borehole and continue drilling to the desired depth. The ocean drilling industry has begun to develop instrumentation for borehole instrumentation where a geophysics package of instrumentation examines the bore hole. Further development along these lines would be invaluable to any drilling that would take place on Mars.

### 8.3.3 Precision sampling of a core

Once a core has been obtained, it would be injected it into a sample triage station. On this station we would like to look down the axis of the core with the contact suite of instruments, especially if the core was delivered intact. This way layering structures can be identified that might be indicative for a sedimentary material or any other area of interest in any type of sample. After the initial analysis, it is not necessarily desirable to process the entire core in an instrument like the rock crusher. If any regions/layers of interest have been identified in the core, these layers would be diluted/mixed with the other less-interesting layers, and thus make the analysis of the core material give results that are not necessarily indicative of a specific region. Furthermore, the rock crushing mechanisms could produce material that is not necessarily representative of the entire core, due to the crushing mechanism. Simply put, a precision sample handling system needs to be developed that is much more advanced then the rock crushers on MSL. This precision sample handling system would most likely replace the rock crushers on MSL as they are currently designed, because there most likely is not enough mass to have both type of instruments aboard.

The precision sample handling system would need to produce fines from a core that are on the order of 100 m, although finer material is always preferable. These fines would need to be produced from regions of the core that are less then 1 cm in length. Although an exact amount of samples that would be useful for each analytical instrument cannot be accurately defined without knowing what instruments have been chosen, a good working number is 100 mg.

A further developed system for AFL would allow for fines to be obtained from various parts of the core, where astrobiologically interesting signatures were observed. These fines then could be identified by the contact suite or by analysis of the fines that were collected during the coring. A possible method this material can be obtained is via a pneumatic drill, analogous to a dentist drill. This type of sample processing of a core needs to be further addressed, as it may require holding the core so that further processing can occur. If the core would be held in a fixed position, a grinder could grind parts of the core while those fines are collected. Any other material that is produced from this step could be looked at with a microscopic imager, if one is chosen as apart of the science payload, but no further analytical analysis of it would need to be made.

### 8.3.4 Ice Samples

Terrestrial organisms can maintain a layer of liquid water around them in an environment where the temperature is below the freezing value for that corresponding pressure. Therefore, one of the main science investigations with ice is to determine if there is any liquid water in a sample. In addition, volatile material present in the sample might undergo reaction and hence change its form, which might lead to incomplete analysis of the obtained sample. Because of these reasons, it seems desirable to obtain a sample without allowing the temperature to pass above conditions in which phase transition that any water (and potential brine solution) present would undergo for the ambient pressure and temperatures present. From permafrost, it is expected that a core approximately 5 cm long and 1 cm diameter would be sufficient for further studies, however, a larger core would likely allow more comprehensive analysis to be performed. With permafrost samples, the possibility should be considered to include a sample concentration device in the sample possessing suite. This concentration process would only take place after any measurement on liquid H2O on the pristine sample has been performed.

The overall strategy for a “polar” mission largely depends on the choice of landing site, i.e. whether it is permafrost or on the polar cap. If a polar region is decided upon, it is suggested that the obtained ice core is analyzed as a whole, especially if only 5 x 1 cm core are obtained. This based on the assumption that the recent ice deposits will not show much evidence of layering or zoning. However, should new data become available through *Phoenix* investigations, this approach may have to be restructured and a more capable precision sub-sampling system be integrated into this mission concept.

### 8.3.5 Liquid and Heat extraction of organics

Organic analysis has been one of the important measurements the Astrobiology Science Steering Group has identified that AFL should be able to perform. In order to identify organic material, they need to be released from the matrix material they are a part of, especially since surface organics on Mars might be very rare. Several ways to extract organics form rock samples were discussed, including using heat and solvent extractions. Each extraction technique has its advantages and disadvantages. However, it is currently not clear whether there would be enough mass to be able to perform both techniques on the same mission. Also, in an extended surface mission (~900 days), which is powered by a nuclear power source, either extraction technique would need to demonstrate that it can perform analysis on a large number of samples (exact number TBD) that such a mission would be able to collect. Different materials require different extraction strategies. For example, some liquid extractions will miss kerogen type material because of its high molecular weight and low solubility, while heat will destroy most of the more fragile biomarkers such as amino acids or hopanes, resulting in the loss of important molecular information. MSL will, most likely, have some form of heat extraction, although what this will look like will be dependent on the instrumentation that is chose for that mission.

Generally, more refractory, fossil, nonpolar compounds require organic solvent extraction. The choice of solvent depends largely on the polarity of the target compounds. Solvents commonly used are for example hexane, dichloromethane, toluoene, methanol, ethylacetate, propanol, or mixtures thereof. It is necessary to identify mixtures that have highest extraction efficiencies and at the same time covering the broadest possible polar-nonpolar stretch. Organic solvents would be needed for GC/MS sample preparation (and to some degree HPLC but this varies depending on column design and target compounds; H2O and methanol are commonly used as eluents, but could also be acetonitrile, dioxane, ethanol, isopropanol, etc.), in order to obtain molecular information from refractory compounds.

Aqueous solvents (such as super critical water) would be used for amino acid, DNA/protein extraction e.g. for microarray analyses, capillary electrophoresis, culturing experiments, flow cytometry or perhaps even PCR. Numerous proprietary and commercially available extraction kits exist using a variety of different solvent compositions, all being aqueous solutions. It would be required to identify optimized procedures and optimized solvents and/or solvent combinations in future laboratory experiments, e.g. using Mars analogue materials spiked with microbial cells and/or organic target compounds.

Other extraction technologies are currently available and need to be examined more in the context of Mars missions these techniques are not limited to but include; microwave, super critical gas (i.e CO2) ultrasonic and sublimation.

It is important to point out here that no judgment is made on which extraction technique is preferable. This is simply an attempt to identify which technique (if either) can be made a facility instrument aboard AFL, and hence have several instruments analyze the extract from the surface samples. Currently, different instrument developers focus on developing extraction techniques for their individual instruments. These techniques do not necessarily have much overlap from instruments to instrument. If consensus could be formed that a particular set of extraction mechanism is desirable (i.e. utilizing H2O at 100oC) it could necessitate a facility instrument to perform that extraction and pass the extract to different instrumentation. This would accomplish reduced mass and power requirements, as well as allow for several instruments to analyze the exact same sample.

Extraction conditions are currently being investigated utilizing different techniques solvents and temperatures. However, the extraction mechanisms need to demonstrate that they are small and repeatable. Null-results from AFL can have great meaning, but they need to be absolute and definable. The Viking GC/MS results showed no organics, but those results don’t necessarily mean there were no organics in the soil that was analyzed. The GC/MS has limits of detection that can be easily determined for single species. However recent work has demonstrated that the Viking ovens were set to a temperature that would have not released certain organics that could have been present in the soil (Glavin et al., 2001). In addition other types of organics could have been destroyed by the heat, and thus not detected. In order to determine what a possible null-result means, an end-to-end analysis would need to be carried out.

Pyrolysis heating:

The Viking landers each had ovens as part of the GC/MS system, although the ovens themselves were not able to reach the temperature necessary to detect some of the organics that could exist there. The Rosetta mission has a small oven, Phoenix has the TEGA, which has eight one-time-only use ovens attached to an EGA and a MS. MSL is intended to investigate multiple samples (24 floor, 78 goal) and has baselined a GC/MS as an instrument. If the development of a multi-use oven is not made, then it would require a ground decision as to whether or not to analyze a sample, if only a limited number of samples that can be analyzed. This would necessitate a science decision, which could delay other analysis on the surface, and limit the number of overall sites that can be visited.

Another prospect of the pyrolysis method is whether it could be designed in such a way that it is capable of concentrating signatures on a sorption trap. If so, any use of those traps will also have to be shown for the same number of samples that the rover will analyze. Also, as mentioned previously, all limits of detection should be for the entire end-to-end system, for a variety of different mineralogical samples.

Liquid extraction:

Liquid extraction is a more gentle way to extract organic molecules from rock and soil samples. One analogy to Martian surface investigations is the analysis of organics from meteoritic material. In those investigations, the organic molecules were released by either hot water extraction or by HCl extraction. Current development of novel techniques for the extraction of exophase biomarkers needs to continue, as does the determining the most efficient solvent extraction parameters. Should a sample be analyzed for its indigenous water content, it might require using another, yet to be determined, technique. In addition, different solvents can extract different types of molecules, water, as it approaches the critical point, becomes a good organic solvent. Clearly, more science groundwork has to be carried out to obtain comprehensive information to allow the best possible choice of solvents to be used. Other solvents that are used in the laboratory include HCl and other acids. These acids perform a more complete digestion of the matrix material, and increase efficiency of extraction, but are harder to handle because of their corrosive nature. With any solvent that is chosen for this step it should be noted that it would be able to concentrate the material to ensure a better signal to noise level.

It is currently unclear whether the liquid sample handling system needs to be completely reusable or whether one-time only use should be the preferred option. This information will become available as experimentation and technology development continues. The only stipulation that needs to be made is that the extraction technique minimizes mass and power resources.

Finally, there are other measurements that can be made during the extraction phase, which would not be possible during pyrolysis heating. These include pH, Redox potential of the material, etc. All of these measurements can help elucidate habitability issues and are an extra measurement that can be made, and if the liquid extraction step is a facility instrument should be made.

### 8.3.6 Contamination concerns

There are two issues that need to be addressed from for contamination concerns:

Contamination issues from organisms brought from Earth

Cross sample contamination

The issue of terrestrial contamination being detected and identified, as material present on a Martian sample is, by far, the main concern. Several different mechanisms can help reduce the possibility of this.

A sterile sample can be brought from Earth and run through the system for the first analysis to show that the end-to-end system is clean and contamination free. If this step produced positive results, it will show that the sample system was not clean and would have to be cleaned e.g. by flushing with a sterile material blank. After the initial sterile material is analyzed, surface dust could be analyzed next. This material is most likely sterile due to UV irradiation and is most likely homogenous across the planet. After the analysis of such sample through the entire system, this material can be used as a negative check for the entire system. If a sample is later found to have the signatures of life, analyzing another soil sample can perform a negative response check of those results, which will further validate the biosignatures that might have been identified.

The other form of contamination is sample to sample. While it should be noted that a general cleaning between samples should be performed, reducing the cross sample contamination should not be a major power and mass drain, which could be better used in other systems.

## 8.4. Time resolved Measurements

For some versions of AFL, time-separated repeat measurements (to observe changes) will be valuable, and these were strongly advocated by some members of the SSG. Given current understanding of Mars, we do not know enough to design the time gap that would be needed in such an experiment (minutes?, hours?, days?, months?), or the fidelity to which the subsequent experiment(s) needs to duplicate the conditions of the first in order to provide a meaningful hypothesis test. The AFL SSG takes the position that time-separated repeat measurements are not essential to all versions of AFL.  Thus, this should not be a part of the common overall mission scientific objectives. The AFL SSG recommends that the capability to do at least some time-separated repeat measurements be a general functionality of the surface science system, and that the decision on how and when to use it be deferred to the competitive process***.***

# 9.0Engineering analysis of AFL core

Based on input from the AFL SSG, a preliminary engineering design concept was defined so that basic mission parameters (such as mass, cost and power generation systems) could be developed. This was done so that technology developments that will be required to undertake the mission could be identified and pursued. This design concept was based upon the AFL SSG core mission requirement and included possible investigation of sedimentary, hydrothermal and liquid water regions. Other investigation (namely to ice covered and sub-surface ice regions) may require a different architecture and hence have a different mass, cost, and power generation systems. The mission architecture was defined by taking into account the measurement objectives, payload infrastructure rover mobility requirements and lander capabilities (Section 8). Given all these requirements and capabilities, a core AFL mission was developed.

The mission studied included 2 instruments for remote sensing placed on the main mast, 2 contact instruments located on an instrument deployment device (IDD), and 6 analytical laboratory instruments capable of analyzing samples obtained from the Martian surface for a total of 10 instruments. The analytical instruments, as well as the sample acquisition and processing infrastructure, will be able to process 25-75 physical samples (rock, regolith, and ice) for detailed analyses by both pyrloysis and wet chemistry instrumentation. Landers, Entry Descent Landing (EDL), cruise launcher, were defined in such a way to meet the mission requirements and so that costing the rover and mission could be done. In order to accomplish this, a list of generic instruments were identified so that parameters such as cost, mass, volume, and power requirements could be included in the engineering design concept. No attempt was made to identify and place individual instruments on the strawman payload (used to assess cost only) and where several instrument from different developers were identified, average mass power and volumes were used.

The engineering design concept assumed a launch in either 2013 or 2018 with a Technology Readiness Level (TRL) of 6 for instruments and subsystem technologies that would have to be reached by 2009 and 2014 respectively. Functional redundancy was required on all subsystems except for the science payload, and this included the sample acquisition and processing infrastructure. Landing site availability for the AFL SSG included access to the Martian surface between: +85 to –60 so that access to both ice regions as well as a wide variety of potential Sedimentary and hydrothermal regions can also be investigated. Landing altitudes of 2.5 km or less relative to the MOLA geoid should be reached within a 10x10 km (3-sigma) landing dispersion ellipse assumed for landing. Because AFL will be assumed to be a mission to a specific site of high scientific interest, rover was designed with “Go-to” mobility capabilities of 10-15 km (linear traverse range) so most astrobiology interesting sites could be reached and explored. For data transmission between Earth and Mars, either MTO or the second generation Mars Telecom Orbiter (MTO) was assumed to be available for Mars to Earth telecom greatly increasing the amount of data that could be acquired on the mission. The collected data would be passed to Earth via 0.3 m HGA for 1024 kbps link via MTO. This design allowed for a data intensive 1-3 GBits of daily science data generation. X-Band from rover direct to Earth (DTE) would be used for back-up purposes only. Finally the main power system of the mission was assumed to be a Radioisotopic Thermal Generator (RTG) system, although solar power could also be utilized for missions that are more equatorial, and potentially shorter in duration (depending on final MER mission power results). The power systems was sized to be able to provide sufficient power with reserves for “worst case” extreme drive Sols (large rocks and slopes) and for analytical laboratory days. Based on this analysis a 4 Brick Small RPS system capable of producing 50We, or 1200WeHRS per sol in combination with a 2 x 8 Ahr-Li-Ion battery system was chosen. Because of the inefficiencies in power generation from an RPS system waste heat has to be dissipated. Therefore, A passive thermal loop system driven by the 1000Wt energy from the RTG system, in combination with electrical heaters, thermal switches, and radiators was designed for the rover for keeping the Warm Electronic Box (WEB), external actuators, and instruments at acceptable temperatures ranges. The passive thermal system on the rover would in combination also be used for dissipating energy from the RTG system the during EDL and cruise stages.

To generate the required science and analyze 25-75 samples, accommodate the selected science payload strawman and provide sufficient power, data storage, data rate, and telecom to an MTO type orbiter, the rover itself would have a mass of ~550 kg (30% reserve included). Of this ~110 kg (~20% of rover mass) would constitute the science payload (once again, depending on the exact parameters of the instruments selected through AO). Bringing such a rover to the Martian surface would require a launched mass of 2456 kg, which would demand an Atlas V521 or a Delta IV 4040 launcher. Assuming, a MER cruise stage, Viking style EDL system with a live lander, this would give an injected mass at Mars of 2174 kg, and require a 4.57 m aeroshell and two chutes during descent.

The rover assumed in this study shares heritage with MER however, final design characteristics for the 2009 MSL mission will influence this decision. The rover includes a mast for the remote sensing instrument, an IDD for the contact instruments and sample acquisition, a detailed sample handling system and an analytical laboratory suite of instruments. The six rover wheels were increased in size to 35 cm (diameter) to negotiate larger rocks and extensive Go-To requirements (as discussed below). Each wheel includes a brushless actuator, which would draw 16-25 W per wheel, and a total of 100-150W for all wheels during traverse depending on surface characteristic of the site (i.e. slop, rock distribution, surface material etc.).

The result of the costing exercise resulted in a 2013 mission cost of $ 1.55 Billion (in RY dollars) and 2018 mission cost of $ 1.78 Billion (in RY dollars). This includes ~ 200 million for instruments and infrastructure and ~ 500 million for all the rover subsystems. These numbers should be adjusted as the design for MSL becomes more set. Savings for things like built-to-print hardware and heritage in the EDL and avionics systems may result in mission savings.

In order to meet the mobility requirements for AFL, the mass of the rover and the potential investigation site are taken into account. One requirement for AFL is to investigate a site(s) that are most likely to have high astrobiology interest. This requirement can mean traverses of up to 10’s of km depending on landing ellipse constraints such that the rover design for longer traverses in Mars terrains must be taken into consideration. In addition, the AFL payload will be much bigger than MER with a scaled rover and hence the wheel contact area has to grow from the 25 cm wheel diameter on MER to accommodate low surface pressure for minimizing wheel sinkage. There are some basic assumptions we can make based upon Mars geology and the proposed investigation sites, such that the mission requirements (see section 8.0) can be accomplished and \ a reasonable preliminary design can be created from which approximate mission costs can be estimated. It needs to be pointed out here that this preliminary analysis is by no means a complete engineering analysis, but it is designed to show approximate system requirements for planning of total mission costs as well as mobility potential for site selection. Finally, with this analysis a decision on the level of required precision (or pinpoint) landing can be made so that investment in technology development for AFL can be carefully planned.

Figure 6. A schematic diagram of how AFL may look



Technology development to fulfill science and engineering goals is summarized in Table 6.

can be seen from Table 6 significant development of critical enabling technology should begin as soon as possible, especially for 2013 launch.

As with all other JPL Rovers, AFL’s drive train subsystem was assumed to be a 6-wheel design. Each wheel has two motors: one turning the wheel, the other steering the wheel. All motors are brushless and 2, 4 or 6 wheels can be driven at a time depending on the terrain. Each wheel consumes approximately 8 W in stand-by mode and about 18 W when driving, making the drive train subsystem the largest power consumer (when operating) on AFL. Additionally, a maximum slope tolerance 30o is assumed due to both current design configurations and projected technology advancement. We have assumed that the technology for continuous drive and autonomous hazard avoidance will be developed and eventually will undergo flight qualification so it can be utilized on AFL. The wheel diameter to be chosen will be large enough to avoid typical Martian hazards (i.e. surface rocks) so that linear odometer distance can be maximized while being small enough to minimize mass and power (which is related to wheel size).

Table 6. Summary of necessary technology for AFL, in particular highlighting instrument development in critical areas as defined by the AFL team. This is not to exclude established technologies from development but merely highlights other critical technologies that should be further developed.

# 10.0Planetary Protection

The different variants of AFL may end up in any of three Planetary Protection classifications.

Category IVb is applied to missions that investigate extant Martian life forms. This may include AFL-Liquid Water and AFL-Ice (depending on the instruments).

Category IVc is applied to missions that access Mars “special regions”. This would include AFL-Liquid Water, AFL-Ice, and perhaps other AFL versions, depending on landing site.

Category IVa is applied to landed missions other than the above. This could apply to AFL-Sedimentary and AFL-Hydrothermal (depending on landing site).

To achieve maximum flexibility, mission engineering should be planned assuming IVb, and de-scoping, if appropriate, can take place from there. The four variants of AFL will have very different implications for Planetary Protection and therefore must be reviewed on a case by case basis.

It is noted that many developing technologies are available for contamination monitoring, decontamination and space craft cleanliness issues. These technologies should be vigorously pursued. In particular the following;

Low temperature sterilization techniques such as microwave plasma and other plasma ashing techniques,

Radiation sterilization technologies for whole space craft as well as ‘hot-spot’ removal.

Real time non culture based systems for monitoring amount and types of bioburden.

Providing of a suitable mineralogical bio and organic clean sample blank for proofing critical sample handling pathways and

It should be noted that several of the analytical techniques mentioned in the AFL instrument section cannot undergo heat sterilization. Protocols that either ensure that instruments are delivered cleaned to the level of the space craft and integrated to the craft after heat sterilization, for the use of cooling loops to keep critical instruments cool during sterilization (obviously the previous point would apply here) or alternatives to heat sterilization must be put in place for these technologies to fly.

# 11.0Relationship between AFL and MSL

AFL will depend on the following heritage from MSL.

Precision landing using a novel (non airbag) landing system

The use of RTG technology

The use of remote, contact and analytical suites of instruments

Crude sample processing to be used but improved on AFL

2.AFL will differ from MSL in the following essential respects:

Advanced sample preparation system.

Precision sub-sampling is an advanced sample management step that will allow a scientific focus on meso- to micro-scale discoveries of enhanced astrobiological interest. This will allow a much higher capacity to investigate specific anomalous features.

Liquid extraction. For advanced studies of carbon chemistry, more efficient sample extraction (and instrument delivery) methods are needed.

Better and miniaturized organic molecule and life-detection related instruments.

Greater interplay between

Precision landing, hazard tolerance/avoidance, go-to mobility.

–Will give us the ability to follow-up on specific discoveries, including in “interesting” terrain.

# 12.0The Future of AFL

It is suggested that the SSG reconvene at a later date to

Respond to discovery to hone mission concepts for site selection

Review sample handling and instrumentation choices and feed-forward to a possible sample return mission

Respond to shifting of the AFL timeline from 2013 to 2018, this would include revisiting the instrument choices based on comments from the SSG as to the use of instruments currently in development but of such a low TRL that it could not feature in the 2013 timeframe example include high vacuum and high voltage instrumentation such as electron microscopy, or photoelectron spectroscopy.

In the past, there was competition between in-situ and sample return mission concepts and there was a question as to whether the AFL was to fly before MSR or after. The current schedule envisions an AFL flight as early as 2016 and an MSR some time after 2020.

The advantages of flying in-situ missions first are that they are relatively low cost compared to MSR (although the costly infrastructure put in place for an initial MSR would not be needed for follow up missions) and there are no issues of sample degradation, sample amount, sterilization, quarantine or ‘off nominal’ delivery to earth.

In addition, the strength of in-situ missions is their ability to assess multiple samples over a spatially diverse area without degradation of the samples. AFL will aid in the identification of sample types for future return missions. This may even include aiding sample caching for a future MSR mission, although that would necessitate a further assessment of precision landing of an MSR mission.

A point to remember is that if / once detected life on Mars should be characterized in its entirety for similarity to earth life, evolution and biochemistry (if viable). Therefore both AFL and MSR must be considered necessary tools to be used at the right time to answer science questions within the foreseeable realms of technology..

Several aspects of both the sample handling capabilities for AFL and the choice of instrumentation will allow the further development of robotic tools to explore elsewhere in the solar system e.g. Europa. This instrumentation although initially geared for the detection of life would upon the successful accomplishment of this task be needed to be further developed to characterize that life in whatever form. It will not be enough to ask was/is there life there, the next logical step is how did it arise, how is it different from earth life and why? It is only by taking this step will we able to understand truly the processes of abiotic / prebiotic / biotic chemistry in the solar system.

Note, the bulk of this work and the draft white paper was completed by September 2004. There have been unavoidable delays to its publication. In the meantime thinking about AFL has progressed. This document reflects the thinking in September 2004. Whilst engineering and programmatic changes have occurred since then, the strength of this document lies in the science definition for the mission.

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# 14.0Appendix 1. Discoveries AFL must respond to.

Table 7 Summarizes crucial science discoveries that may also directly affect AFL mission, potential follow up questions and measurements

Generated

# 15.0Appendix 2 - Instrument descriptions and capabilities

In this table, a number of techniques were suggested by AFL SSG members as potentially applicable to one or more identified measurement objectives. This list is not meant to be comprehensive or definitive, but rather to illustrate the kinds of information that would enable instrument development efforts in general to connect to the specific needs of AFL. As such the table does not identify all aspects of each technique, but only those that were discussed in a preliminary analysis of the desired measurements on AFL. The first and second columns identify the technique and the type of measurement(s) with which it is typically associated (Data/Signatures Sought). The third column explicitly lists the most likely AFL measurement requirement that the technique addresses (see Section 7.0). In this way, techniques applicable to a given measurement of interest, or more generally to a mission objective (see Figure 5), can be found by examining those rows containing the category (1-5) desired. This column is meant to serve as an example template, so all potential uses of each technique are not identified. The next three columns indicate the most likely associated tier(s) for the technique, corresponding to the recommended division as discussed above.

The following thirteen columns provide data for *example* implementations of the technique where useful specifications of the sample analyzed and typical instrument parameters could be identified. Given sample data include: 1. the physical form *as acquired or as extracted/analyzed* – solid (s), liquid (l), or gas (g); 2. the type of material from which it is obtained and/or delivered to the instrument; 3. the type of sample preparation required and/or desired (see key); and 4. the typical size or mass of sample, additionally indicating where a technique looks only at the surface of a solid sample rather than the bulk. The first three columns of the Example Technique Characteristics section provide some of the key distances involved: the standoff, the field-of-view (FOV) or spot size, and the scale of the heterogeneity probed, if appropriate. The heterogeneity is indicated by the structures (e.g., layers or grains) that can be individually analyzed with the method’s FOV or spot size. For example, a Hand Lens instrument might look at individual mineral grains and similar size structures within a mm-cm FOV from a standoff focal length of a cm or so. In this example it is the imaging resolution, not the FOV, that determines the smallest structures observable, and that additional data is found in the resolution column. On the other hand, for a laser mass spectrometer, the spot size does roughly determine the spatial resolution of analysis – a spot size below 100 microns could enable analyses of mineral phases on the mm scale; what is then found in the resolution column is in fact the mass resolution, since that is how the term is used for that method. Further, the Mass Range column gives the typical range of molecular weights that are accessible with a given mass spectrometric method.

Finally, the remaining columns provide a correlation of where a technique would be applied in support of various *discovery-responsive measurements* by AFL that would be called for following the discoveries listed in Appendix 2. This separate correlation, beyond the technique-to-measurement requirement-to-mission objective logical chain, permits a greater flexibility and responsiveness of the AFL concept to specific scenarios that may develop from current Mars missions and over the next several years.

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Table 8- Techniques Suggested for AFL by SSG Members** | | | | | | | | | | | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  | Measurement Tier | | | Example Target/Sample Information | | |  | Example Technique Characteristics (instrument implementation) where appropriate | | | | | | | |  | Discovery/Follow-up per Table 7 | | | |  |  |  |
| Technique | Data/ Signatures Sought | Mmnt Reqts Addessed (Section 7) | Remote Sensing/Standoff | Contact or Close Range | Analytical Lab | Physical Form (Solid, Gas, Liquid) | Example Origin/Host Material | Processing Required/ (Desired) | Sample Mass/ Volume | Distance to Target | Size of Area Probed/ FOV | Target Feature Scale | Selectivity | Detection Limits | Resolution | Precision | Mass Range | Other | Recent Surface Water | Hydrous Mineral Phases | Organic Molecules | Sedimen. Structures | Sedimen. Rocks | Evidence for Fossil Life | Microbes |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| stereo optical imaging | identify targets, evidence of weathering, sedimentation, alteration, etc. | A | x |  |  |  | sedimentary rocks/ structures | n |  | 1m - 10+km | 10cm - 1+km |  |  |  |  |  |  |  | 1 | 6 |  | 1,2,3,4 | 4 | 1,2,3,6,8 | 3 |
|  | identify surface samples | A |  |  |  |  |  |  |  | 10-100 m | 1-10 m | 10cm-1m |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | identify distant sedimentary outcrops | A |  |  |  |  |  |  |  | 1 km | 10-100 m | 1-10 cm |  |  | 10 cm @ 1 km | |  |  |  |  |  |  |  |  |  |
| VIS/NIR Spectroscopy | surface mineralogy, texture | B | x | x |  | s | rocks, fines | n (abr) |  | cm - m |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| mini TES | mineralogy | B | x |  |  | s |  | n (abr) |  | m - km |  |  |  |  |  |  |  |  |  |  |  |  | 1 |  |  |
| long focal length imaging | identify distant sedimentary outcrops | A | x |  |  |  |  | n |  | 10m - km | cm-10m | cm |  |  |  |  |  |  |  |  |  |  |  |  |  |
| laser ranging | distance to target | A | x |  |  |  | boulders, vertical faces | n |  | 100m - km | cm spot |  |  |  | cm @ 100 m | |  |  |  |  |  |  |  |  |  |
| LIBS | elemental composition | B | x |  |  | s | boulders, slopes | n |  | 1 - 25m | mm - cm spot |  | low (l absorb.) | ppmw |  | ~ 10% | elements | laser ablation | | 2,4 |  |  | 2,5 | 3 |  |
| ground penetrating radar | ice, H2O, other | B, C | x |  |  | s | subsurface | n |  | m - 10s m |  |  |  |  |  |  |  |  | 3 |  |  |  |  |  |  |
| seismic sounding | ice, H2O, other | B, C | x | x |  | s | subsurface | n |  | 100's m - km |  |  |  |  |  |  |  |  | 3 |  |  |  |  |  |  |
| neutron spectroscopy | ice, hydrated minerals | B | x | x |  |  | drill cores, fines | n (acq) |  | 10's cm - m's |  |  | high | variable <%-% | |  |  |  |  |  |  |  |  |  |  |
| gamma ray spectroscopy | elemental composition | B | x | x |  |  | any | n |  | 10's cm - m's |  |  | med | variable <%-% | |  |  |  | 5 |  |  |  |  |  |  |
| x-ray spectroscopy | elemental composition | B, E2 |  | x |  |  | any | n (acq) |  | cm | cm+ |  | med | variable <%-% | |  |  |  |  |  |  |  |  |  |  |
| Raman spectroscopy | mineralogy, some geochemical/organic | B, E | x | x |  |  | rocks | n (abr) |  | cm - m | cm+ |  | med |  |  |  |  |  |  | 1,5 | 1,4 |  | 2 | 1 |  |
| micro-Raman spectroscopy | mineralogy, some geochemical/organic | B,D,E |  | x | x |  | rock chips | n (acq, abr) | | mm - cm | < mm |  | med |  |  |  |  |  |  | 1,5 | 1,4 |  | 2 | 1 |  |
| micro-LIBS | elemental composition | B,D |  | x | x | s | rocks, chips | acq,pos |  | mm - cm | < mm |  | low (l absorb.) | |  |  |  |  |  | 2,4 |  |  | 2,5 | 3 |  |
| hand-lens-scale imaging | phase texture/identity | D |  | x |  | s |  | n (abr) |  | cm - m's | 0.1-10 mm | grains |  |  |  |  |  |  |  |  |  |  |  |  |  |
| optical microscopy | fine morphology | D,E |  | x | x | s |  | n (abr) |  | mm - cm | 0.001-1 mm | subgrain |  |  |  |  |  |  |  |  | 2 |  |  | 1,2,3,6,8 |  |
| confocal microscopy |  | D,E |  |  | x | s |  |  |  | mm | 0.001-1 mm | subgrain |  |  |  |  |  |  |  |  | 2 |  |  | 1,2,3,6,8 |  |
| near-field microscopy | very high res imaging | D,E |  |  | x | s | flat chip | acq, pos |  |  |  | subgrain |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Mossbauer | Fe-bearing mineralogy | B |  | x | x | s |  |  |  | mm - cm |  | avg |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Fe-NMR |  | B |  | x |  | s |  |  |  | mm - cm |  |  | high |  |  |  |  |  |  |  |  |  |  |  |  |
| XRD/XRF | mineralogy | B,D |  |  | x | s | drill cores, fines | acq, pow | mg's | 0 | whole sample | avg or grains | |  |  |  |  |  |  | 1 |  |  | 1,5 |  |  |
| FTIR | mineralogy, some geochemical/organic | B |  |  | x | s |  |  |  |  |  |  |  |  |  |  |  |  |  | 1,5 | 1,4,6 |  | 2,5 | 3 | 4 |
| VCD |  |  |  |  | x | s |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| deep UV fluorescence | organics: identity, oxidation state, … | B,D,E |  | x | x | s |  | n (abr) | surface? | mm - m |  | grain scale+ | |  |  |  |  |  |  |  | 1 |  |  | 3 |  |
| pyrolysis/GCMS | organic and some mineralogical/inorganic composition; isotopes | B,E |  |  | x | s,g,l |  | acq, pos, vac | mg-10's mg | 0 | whole sample | avg | low |  |  |  |  |  | 4 |  | 1,5 |  | 5 |  |  |
| chemical derivatization | less-tractable organics | E |  |  | x | s,l |  | liq |  | 0 | whole sample | avg |  |  |  |  |  |  | 4 |  | 1,4,7 |  | 5 |  |  |
| isotope ratio MS (IRMS) | C and other isotopes for bio-fractionation, age dating | E2 |  |  | x | s,l |  | acq, pos, vac | | 0 | whole sample | avg |  |  |  |  |  |  | 2 |  |  |  |  |  |  |
|  | compound-specific IRMS using sampling selectivity | E2 |  |  |  |  |  |  |  |  | whole sample | avg | cmpd isolated w/pyr, GC,or other proc. | | | |  |  |  |  |  |  |  |  |  |
| chiral GC | enantiomeric excess (ee) | E3 |  |  | x | s,g,l |  | acq, pos, gas | | 0 | whole sample | avg |  |  |  |  |  |  |  |  | 3 |  |  |  |  |
| circular dichroism | enantiomeric excess (ee) | E3 |  |  | x |  |  |  |  | 0 |  | avg |  |  |  |  |  |  |  |  | 3 |  |  |  |  |
| liquid chromatography (LC) | organics, ee | E |  |  | x | s,l |  | liq |  | 0 | whole sample | avg |  |  |  |  |  |  | 4 |  | 1,3,5 |  | 5 |  |  |
| 2D GCMS/TOF-MS | organic and some mineralogical/inorganic composition; isotopes | B,E |  |  | x | s,g,l | rocks/cores, fines | acq, pos, vac | 10's mg | 0 | whole sample | avg | low |  |  |  | ~1E3-1E5+ | | 4 |  | 1,5 |  | 5 |  |  |
| electrospray ionization MS (ESI/IMS/CIT-MS) | | E |  |  | x | s,g,l |  | acq, pos, vac | g's | 0 | whole sample | avg | low |  | Dm/m 1E2-1E3+ | |  | contact w/ fluidized sample | | |  |  |  |  |  |
| laser ablation TOF-MS | local elemental/isotopic composition | B,D |  |  | x | s | rock chips, fines | acq, pos, vac | surface | 0 | 10mm - 1 mm | grain scale+ | low (l absorb.) | ppbw-ppmw | Dm/m 1E2-1E3 | 5-25% | ~ 300 |  | 4 | 2,4 |  |  | 2,5 | 1,3 |  |
| LD/MALDI-TOF MS | high-MW organics; some inorganic molecules | D,E |  |  | x | s | rock chips, fines | acq, pos, vac (pow, liq) | surface/prep film | 0 | 100mm - 1 mm | grain scale+ | med (l absorb.) | fmol-pmol | Dm/m 1E3-1E4 | | ~1E3-1E5+ | |  |  | 1,2,4,7 |  |  | 3 | 4 |
| REMPI-MS/RIMS | organics, elements (trace) | E |  |  | x | s |  | acq, pos, vac | | 0 | 10mm - 1 mm |  | very high (l absorb.) | s. atom - pmol | Dm/m 1E2-1E4 | | ~ 1E3 |  |  |  |  |  |  |  |  |
| AP-MALDI-MS (TOFMS or ITMS) | organic, inorganic molec. | D,E |  | x | x | s | rocks, ices | vac | surface | mm | 10mm - 1 mm | grain scale+ | med (l absorb.) | fmol-nmol | Dm/m 1E3-1E4+ | | ~ 1E3-1E5 |  |  |  |  |  |  |  |  |
| electrospray TOF-MS | high-MW organics | E |  |  | x | s,l | rocks/cores, fines | acq, liq, pos, vac | | 0 | whole sample | avg | med |  | Dm/m 1E2-1E4 | | ~1E4-1E5+ | |  |  | 1,2,7 |  |  | 3 | 4 |
| TOF-SIMS | chemical imaging | B,E |  |  | x | s | rock chips | acq, pos, vac | surface | 0 | 50nm-50mm | sub-grain+ | low |  | Dm/m 1E3-1E4 | | ~1E3-1E4 |  |  |  |  |  |  |  |  |
| ICP-MS | trace elements | B |  |  | x | s,g,l | rock chips, fines | acq, pos, vac, gas | | 0 | whole sample | avg or grains | low | pptw-ppbw | Dm/m 1E3+ | 0.1-10% | ~ 300 |  | 2,4 |  | 1,3,5 |  | 5 |  |  |
| TIMS | isotope ratios (~IRMS) | B,E2 |  |  | x | s |  | acq, pos, vac | | 0 | whole sample | avg | low | pptw-ppbw | Dm/m 1E3+ | 0.1-1% | ~ 300 |  | 2 |  | 5 |  |  |  |  |
| AFM | nanoscale imaging | D, E3 |  |  | x | s | flat chip | acq, pos | chips |  | 1nm-1mm | sub-micron | |  |  |  |  |  |  |  |  |  |  |  |  |
| TEM/SEM | nanoscale imaging | D |  |  | x | s | flat chip | acq, pos, vac | chips |  | 1nm-1mm | sub-micron | |  |  |  |  |  |  |  |  |  |  |  |  |
|  | image microbes in ice cores | D,E3 |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| XPS | chemical comp. and bond state | B,C |  |  | x | s,l |  | vac | 100's mg |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Auger spectroscopy | bond state of elements | B,C |  |  |  | s,l |  | vac | 100's mg |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| amino-acid sensors (eg MOD) | detection of amino acids | E |  |  | x | s,l |  | acq | 100's mg |  | whole sample |  | high |  |  |  |  |  |  |  | 1,3 |  |  |  |  |
| oxidant sensors | detection of oxidants | C |  |  | x | s,l |  | Acq, dry | 100's mg |  | whole sample |  | high |  | per sample weight | |  |  |  |  | 6 |  |  |  |  |
| bio-assay chip lab |  | E |  |  | x | s,l |  | liq | 100's mg | 0 | whole sample |  | high | pptw | per sample weight | | Kda |  |  |  | 7 |  |  | 1,5,6 | 1,5,6 |
| micro-array sensors |  | E |  |  | x | s,l |  | Liq | 100's mg |  | whole sample |  | high | pptw | per sample weight | | Kda |  |  |  | 7 |  |  | 1,5,6 | 1,5,6 |
| MORD |  |  |  |  | x | s |  |  | 100's mg | 0 | whole sample |  | high |  | per sample weight | |  |  |  |  |  |  |  |  |  |
| fluorescence staining | organics | E |  | x | x | s,l |  | Liq | 100's mg |  | whole sample | avg | high | single cell | per sample weight | |  |  |  |  | 7 |  |  |  |  |
|  | SYBR gold, SYTO, DAPI nucleic acid stains for counting microbes | E |  |  | x | s,l |  | Liq | 100's mg |  | whole sample |  | Medium | single cell | per sample weight | |  |  |  |  |  |  |  |  | 1.5.6 |
|  | CTC, tetrazolium salt redox stains for individual cells | E |  |  | x | s,l |  | Liq | 100's mg |  | whole sample |  | Medium | Single cell | per sample weight | |  |  |  |  |  |  |  |  | 1,5,6 |
| isotopic labelling | 14CO2 or 3H for total population activity | E |  |  | x | s,g,l |  | Lig | 100's mg |  | whole sample | avg | medium | single cell | per sample weight | |  |  |  |  |  |  |  |  | 1,5,6 |
| flow-cytometry |  | E |  |  | x | s,l |  | liq | 100's mg |  | whole sample | avg | medium | single cell | per sample weight | |  | If have required media | | |  |  |  |  | 1,5,6 |
| culturing/cell-growth assays |  | E |  |  | x | s,l |  | liq | 100's mg |  | whole sample | avg | high | single cell | per sample weight | |  | If have required media | | |  |  |  |  | 1,5,6 |
| ATP and LAL enzyme assays |  | E |  |  | x | s,l |  | liq | 100's mg |  | whole sample | avg | high | pptw | per sample weight | |  |  |  |  |  |  |  |  | 1,5,6 |
| DNA extraction/PCR |  | E |  |  | x | s,l |  | liq | 100's mg |  | whole sample | avg | high | 100 cells | per sample weight | |  | with correct primers | | |  |  |  |  | 5,6 |
| capillary electrophoresis (CE) |  | E |  |  | x | s,l |  |  | 100's mg |  | whole sample | avg | high | pptw | per sample weight | |  |  |  |  | 7 |  |  |  | 1,5,6 |
| microcalorimetry |  |  |  |  | x |  |  |  | 100's mg |  | whole sample | avg | medium | pptw | per sample weight | |  |  |  |  | 7 |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| **KEY:** |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| n - can be operated with no sample acquisition/processing | | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| abr - abrasion to remove surface layers | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| acq - sample acquisition from host matl (via whatever means) | | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| pow - powdering of solid sample | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| pos - sample positioning (e.g., manipulation to oven, point of focus or extraction) | | | | | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| vac - vacuum processing |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| liq - liquid processing |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| gas - gas processing |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |