Friday, October 17, 2003
Afternoon Session I
SPECIAL SESSION IN MEMORY OF DAVID WYNN-WILLIAMS: LIFE AND ITS DETECTION IN EXTREME POLAR ENVIRONMENTS *(Continued)*
1:30 p.m. Victoria Room

Doran P. T. * Bar-Cohen Y. Fritsen C. Kenig F. McKay C. P. Murray A. Sherrit S.
Life Detection and Characterization of Subsurface Ice and Brine in the McMurdo Dry Valleys Using an Ultrasonic Gopher: A NASA Astep Project **[#8019]**

Fritsen C. H. * Priscu J. C. Doran P. T.
Bacterial Distribution and Production Within Lake Ice and Glacial Ice Along the Fringe of the Antarctic Ice Cap **[#8120]**

Christner B. C. * Priscu J. C.
Earth's Icy Biosphere **[#8121]**

Becker L. * Brinckerhoff W. Cotter R. J.
Detection of Organic Compounds in Polar Ices on Mars Using AP MALDI **[#8122]**

PANEL DISCUSSION
FINDING EVIDENCE OF LIFE IN ICY ENVIRONMENTS
Panelists: TBD

GENERAL DISCUSSION

3:30 – 3:45 BREAK

LIFE DETECTION AND CHARACTERIZATION OF SUBSURFACE ICE AND BRINE IN THE MCMURDO DRY VALLEYS USING AN ULTRASONIC GOPHER: A NASA ASTEP PROJECT.

P. T. Doran[1], Y. Bar-Cohen[2], C. Fritsen[3], F. Kenig[1], C. P. McKay[4] A. Murray[3] and S. Sherrit[2] [1]University of Illinois at Chicago, Earth and Environmental Sciences, 845 West Taylor Street (MC186), Chicago, IL 60607 USA email:pdoran@uic.edu or fkenig@uic.edu, [2]CALTECH, Jet Prop Lab, 4800 Oak Grove Dr, M-S 82-105, Pasadena, CA 91109 USA email: yosi@jpl.nasa.gov or ssherrit@jpl.nasa.gov, [3]Desert Research Institute, 2215 Raggio Parkway, Reno, NV 89512 USA email:cfritsen@dri.edu or alison@dri.edu, [4]NASA Ames Research Center, Divion of Space Science, Moffett Field, CA 94035 USA, email:cmckay@mail.arc.nasa.gov

Introduction: Evidence for the presence of ice and fluids near the surface of Mars in both the distant and recent past is growing with each new mission to the Planet. One explanation for fluids forming spring-like features on Mars is the discharge of subsurface brines. Brines offer potential refugia for extant Martian life, and near surface ice could preserve a record of past life on the planet. Proven techniques to get underground to sample these environments, and get below the disruptive influence of the surface oxidant and radiation regime, will be critical for future astrobiology missions to Mars. Our Astrobiology for Science and Technology for Exploring Planets (ASTEP) project has the goal to develop and test a novel ultrasonic corer in a Mars analog environment, the McMurdo Dry valleys, Antarctica, and to detect and describe life in a previously unstudied extreme ecosystem; Lake Vida (Fig. 1), an ice-sealed lake.

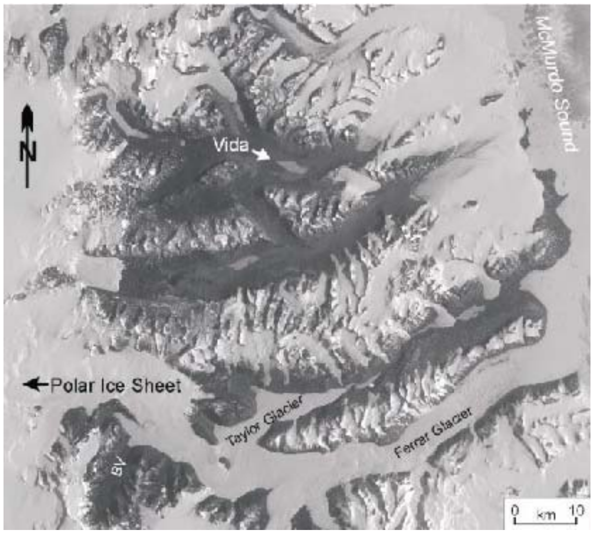

Figure 1: Landsat image of the dry valleys region showing location of Lake Vida. The image is centered at 77.5oS 162oE.

Ice-Sealed Lakes: Lakes in the McMurdo Dry Valleys of East Antarctica have long been studied as extreme environments and potential analogs of purported Martian lakes of the past [e.g. 1, 2]. Commonly studied dry valley lakes have a 2 to 6 m perennial ice cover and 20 to 60 m water column beneath. These lakes also have a range a salinities from fresh to hypersaline, and all allow suffienct sunlight to pass through the ice for photosynthesis to occur in the water column and benthos.

A few lakes in the dry valleys have been largely unstudied until recently because they were believed to be frozen to their beds. One of these lakes (Lake Vida) is also one of the two largest lakes in the dry valleys. Using a combination of ground-pentrating radar and ice coring techniques we have established that Lake Vida comprises a NaCl brine with a salinity seven times sea water and temperature constantly below -10°C lies beneath ~20 m of ice that is at least 2,800 radiocarbon years old [3]. Microbial mats occur throughout the ice column and are viable upon thawing. Sediment layers in the ice effectively block incoming solar radiation (Fig. 2).

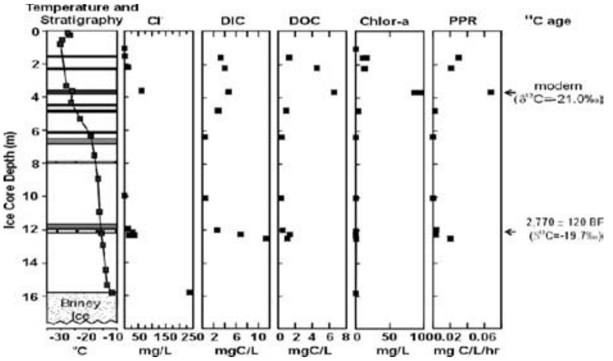

Figure 2: Physical and chemical properties of Lake Vida ice core taken in October 1996. Black horizons on the stratigraphy plot represent sediment layers, gray horizons are sandy ice, and vertically banded horizons contain microbial mat. The temperature profile shown was taken at the time of ice extraction.

Ultrasonic Gopher: Planetary sampling using conventional drilling and coring techniques is limited by the need for high axial force necessitating the use of heavy rovers or anchoring mechanisms. A novel ultrasonic/sonic driller/corer (USDC) mechanism [4] was developed that overcomes these and other limitations of conventional techniques. The novel element of the USDC is the free-mass that operates as a frequency transformer converting 20 KHz ultrasonic waves to a 60-1000 Hz sonic hammering action (percussion) that is applied onto the drilling/coring bit. The USDC actuator consists of a stack of piezoelectric ceramics with a backing material that focuses the emission of the acoustic energy forward, and a horn that amplifies the displacements generated by the stack. The tip of the ultrasonic horn impacts the free-mass creating a sonic resonance between the horn and the bit.

The USDC has been demonstrated to drill rocks that range in hardness from hard granite and basalt to soft sandstone and tuff. This novel drill is capable of high-speed drilling (2 to 20 mm/Watt·hr for a 2.85mm diameter bit) in basalt and Bishop Tuff using low axial preload (<10 N) and low average power (<5 W). The USDC mechanism has also demonstrated feasibility for deep drilling. The Ultrasonic-Gopher (Fig. 3a) can potentially be used to reach great depths and large diameters (3 and 4.5 cm have been demonstrated) using a low mass rover. Generally, the bit creates a borehole that is larger than the drill bit outer diameter and it also creates a core that is smaller in diameter than the inner diameter of the coring bit. This reduces the chances of bit jamming where hole integrity is maintained, and it eases in the extraction of the core from the bit. Current models suggest that the USDC performance does not change significantly with changes in ambient gravity.

Figure 3: a) Ultrasonic-Gopher and extracted limestone core, b) Recent prototype of ice gopher. Ice chisel bit on the left and actuator on the right.

"Ice Gopher": Ice below 16 m depth in Lake Vida and the brine body have never been sampled directly due to logistical constraints. We are building an ultrasonic "ice gopher" (Fig. 3b) to make in situ ecosystem measurements, and acquire samples to be further analyzed. Early versions of the "ice gopher" suggest that coring through ice may prove a bigger challenge than coring through rock. A large part of our efforts in the early stages of development are focused on the problems of chip handling and ice melt during drilling, both of which can create significant potential for getting the instrument stuck during the mission.

Our field plan is to use the gopher to core through the Lake Vida ice cover, cycling in and out of the hole to retrieve ice cores along the way. The gopher will sample brine as it goes and the brine will be collected at the ice surface under clean and sterile conditions. Using the gopher we will address two main hypotheses

H1. Microbial communities within the brine (include brine pockets in the deep ice) and benthic sediments are currently viable, active and affect the present-day geochemistry of the lake.

H2. The ice, water column and benthos of deeply frozen lakes contain geochemical signatures of past microbiological activity.

Conclusions: Lake Vida provides the unique opportunity to investigate lake ecosystems on the edge of existence to determine what conditions may lead to the eventually complete freezing of a lake and the subsequent development/evolution of microbial communities and geochemical signatures. The combined hypersaline, aphotic, atmospherically isolated and cold conditions in Lake Vida make it potentially among the most extreme aquatic environments on Earth. These conditions were likely to have been present during the last stages of purported lakes on Mars near the end of its water-rich past. Our drilling program will provide useful insight into the challenges of drilling though cold dirt and ice with a low power and light weight instrument to retrieve samples for life detection.

References: [1] McKay C. P. et al. (1985) *Nature, 313,* 561-562. [2] Doran P. T. et al. (1998) *JGR, 103(E3),* 28481-28493. [3] Doran P. T. et al. (2003) *PNAS 100,* 26-31. and Author C. D. (1997) *JGR, 90,* 1151–1154. [4] Bar-Cohen Y. et al. (2001) "Ultrasonic/Sonic Driller/Corer (USDC) With Integrated Sensors," U.S. Patent filed.

BACTERIAL DISTRIBUTION AND PRODUCTION WITHIN LAKE ICE AND GLACIAL ICE ALONG THE FRINGE OF THE ANTARCTIC ICE CAP.

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Introduction: Microbial growth within Earth's icy habitats can help define where microbial consortia may survive and function within similar environments on other planets.

Herein, we report on ice properties, bacterial biomass and rates, of bacterial production within perennial lake ice covers and glacial ice environments within the McMurdo Dry Valleys, Antarctica which lie at the border of the Antarctic polar ice cap along the Victoria land coast (Fig. 1).

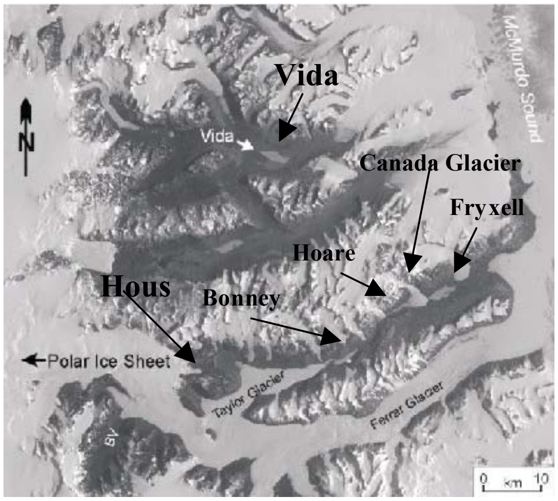

Figure 1: Landsat image of the McMurdo dry valleys region showing location of Lakes Vida, Hoare, Fryxell, Bonney as well as the Taylor, and Canada glaciers. The image is centered at 77.5^{o}S 162^{o}E.

Permanently Ice covered Lakes: Lakes in the McMurdo Dry Valleys of East Antarctica have long been studied as extreme environments and potential analogs of purported Martian lakes of the past [e.g. 1, 2]. Typical, Dry Valley lakes have a 2 to 6 m perennial ice cover overlying 20 to 60 m water columns. These lakes have a range a salinities from fresh to hypersaline, and all allow sufficient sunlight to pass through the ice for photosynthesis to occur in the water column and benthos. Some lakes (e.g. Vida and House) have much thicker ice covers, and it is unknown if the water pockets beneath these ice covers contain viable microbial communities.

Sediment-microbial consortia which are invariably associated with bubble features indicative of liquid water pockets (Fig. 2) are found within the cold ice covers of these lakes [3].

Figure 2. Sediment inclusion from Lake Bonney, with associated arching bubbles indicating the past presence of a melt water pocket. Scale bar is approximately 10 cm.

Glacial Cryoconites: Ablation zones of glaciers also contain sediment inclusions (cryoconites) that harbor microbial consortia [e.g. 4,5]. The sediment-microbiota inclusions are associated with clearer ice created by the melting and refreezing processes which metamorphose the opaque glacial ice by dispersing the small glacial air pockets.

Figure 3: Cryoconite on the Canada Glacier next to author's mitten.

Bacterial numbers: Bacterial cell concentrations range from 4.9e4 to 1.9e7 cells per ml of ice melt water in the lake ice and glacial ice environments. The higher concentrations of bacterial biomass are associated with the ice where sediment inclusions and metamorphosed ice and air inclusions exist (Fig. 3).

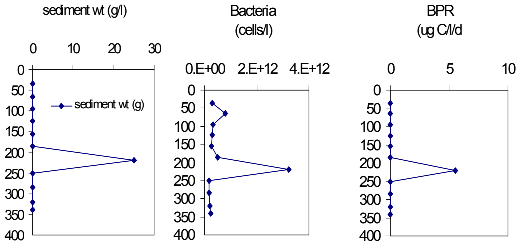

Figure 4. Profiles of sediment content, bacterial cells and bacterial production in the ice cover of Lake Hoare. The coincident peaks of sediment and bacterial abundance and activities are representative of profiles in other lakes as well as enhanced activities in glacial ice with sediment inclusions.

Bacterial Production: Bacterial production rates (BPR) ranged from below levels of detection to 21 ug C l^{-1} d^{-1} in ice melt water. Rates in ice with sediment inclusions averaged ca. 1 ug C l^{-1} d^{-1}.

Rates in ice melt water are not necessarily indicative of *in situ* rates. However, during the summer, these ice habitats experience radiation-induced internal melting [e.g. 6, 7] that creates ice melt water microenvironments. Hence, rates in melt water may be indicative of ice-bound processes. Interestingly, rates of bacterial production relative to measured rates of primary production (PPR) (primarily by cyanobacteria [8]) were comparable (exhibiting rates close to unity) in the samples where BPR and PPR both exceeded 0.1 ug C l^{-1} d^{-1} (Figure 5). Such coincidence in the magnitude of production may be indicative of coupled successional feedback processes that are expected within enclosed systems that reach near steady-state conditions. If these environments do indeed exhibit close coupling we would expect that biomass accrual and biosignature development will be tightly coupled to the ice dynamics that would provide new material to these habitable zones (also see [9]). Recognition of morphological features on the martian polar icescape indicative of such processes may be key to the search for habitable microzones on the Red (and white) Planet.

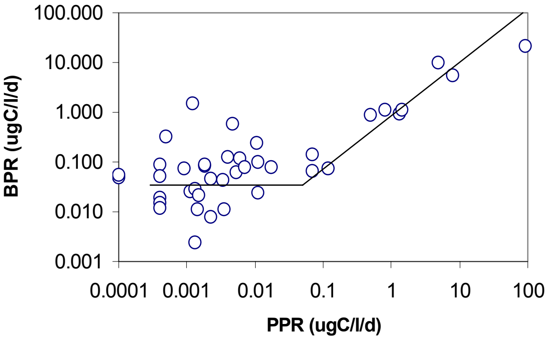

Figure 5: Rates of bacterial production (BPR) relative to primary production (PPR) within lake ice and glacial ice of the McMurdo Dry Valleys.

References: : [1] McKay C. P. et al. (1985) *Nature, 313,* 561-562. [2] Doran P. T. et al. (1998) *JGR, 103(E3),* 28481-28493. [3] Priscu, J.C. et al. (1998) *Science*, 280, 2095-2098. [4] Christner et al. (2002) *Extremophiles*. [5] Muellor D.R. et al. (2001) *Nova Hedwigia*, 123, 173-197. [6] Fritsen, C.H. et al. (1998) *Antarctic Research Series,* 72:269-280. [7] Adams, E.E. et al. (1998) *Antarctic Research Series,* 72:255-268. [8] Fritsen, C.H. et al. (1998) *J. Phycol,* 34, 587-597. [9] Priscu and Christner (In press), *Microbial Diversity and Prospecting.*

EARTH'S ICY BIOSPHERE.

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Abstract: Earth's biosphere is cold, with 14% being polar and 90% (by volume) cold ocean <5 ºC. More than 70% of Earth's freshwater occurs as ice (Fig. 1) and a large portion of the soil ecosystem (~20%) exists as permafrost.

Figure 1. Global locations of existing glacial ice sheets and caps (denoted by shading).

Paleoclimate records for the past 500,000 years have shown that the surface temperature on Earth has fluctuated drastically, with strong evidence showing that the Earth was completely ice-covered during the Paleoproterozoic and Neoproterozoic periods [1, 2]. New discoveries of microbial life in cold (-5ºC) and saline lakes, permanent lake ice, cryoconite holes, polar snow, glacial ice, and subglacial environments are extending the known boundaries of the biosphere. Despite the mounting evidence for microbial life in frozen ecosystems, little is know about the psychrophilic or psychrotolerant microorganisms that inhabit them. Molecular-based ecological studies have revealed close phylogenetic relationships between isolates from global locations, with little in common between these environments except that all are permanently cold (Fig. 2), arguing that microorganisms from these genera evolved under cold circumstances and likely possess survival strategies to survive freezing and remain active at low temperature.

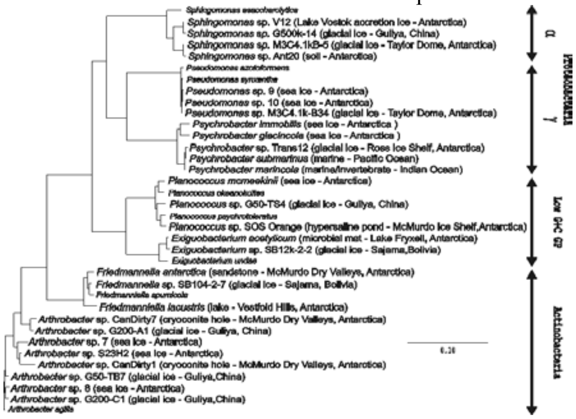

Figure 2. Neighbor-joining tree based on 16S rDNA sequences obtained from isolates (shown in bold) inhabiting permanently cold environments.

Studies of Earthly ice-bound microbes are also relevant to the evolution and persistence of life on extra-terrestrial bodies. During the transition from a clement environment to an inhospitable environment on Mars, liquid water may have progressed from a primarily liquid phase to a solid phase and the Martian surface would have eventually become ice-covered [3]. Habitats in polar ice may serve as a model for life on Mars as it cooled and may assist us in our search for extinct or extant life on Mars today. Biochemical traces of life or even viable microorganisms may well be protected from destruction if deposited within polar perennial ice or frozen below the planet's surface. During high obliquity, increases in the temperature and atmospheric pressure at the northern pole of Mars could result in the discharge of liquid water that might create environments with ecological niches similar to those inhabited by microorganisms in terrestrial polar and glacial regions. Periodic effluxes of hydrothermal heat to the surface could move microorganisms from the martian subterranean, where conditions may be more favorable for extant life. The annual partial melting of the ice caps might then provide conditions compatible with active life or at least provide water in which these microorganisms may be preserved by subsequent freezing [4].

We propose that the Earth's cryosphere and associated sub-ice lakes should be included as biospheric components of our planet. Here, the cryosphere is defined as that portion of the Earth's surface where water is in a solid form as snow or ice, including solid forms such as sea ice, freshwater ice, snow, glaciers, and frozen ground. Examining permanently ice-covered habitats and microorganisms preserved for extended periods within ice is relevant to astrobiological discussions of past or present life on Mars, and the concept that planetary bodies may not be biologically isolated. Such remote and seemingly inconsequential frozen environments may harbor as yet undiscovered microbial ecosystems that could shed light on the natural history and evolution of life on a frozen Earth, as well as other icy planets and moons in the solar system.

References: [1] Kirschvink, J.L., E.J. Gaidos, L.E. Bertani, N.J. Beukes, J. Gutzmer, L.N. Maepa, and R.E. Steinberger. (2000) *PNAS*, 97, 1400-1405. [2] Hoffman, P.F., A.J. Kaufman, G.P. Halverson, and D.P. Schrag. (1998) *Science,* 281, 1342-1346. [3] Wharton, R.A., Jr., R.A. Jamison, M. Crosby, C.P. McKay, J.W. Rice, Jr. (1995) *J. Paleolimnology,* 13, 267-283. [4] Clifford, S.M. et al., (2000) *Icarus,* 144, 210-242.

Detection of Organic Compounds in Polar Ices on Mars Using AP MALDI Luann Becker[1], William Brinckerhoff[2] and Robert J. Cotter[3], [1]Department of Geology, University of California, Santa Barbara, 1140 Girvetz Hall, Santa Barbara, CA 93106 email: lbecker@crustal.ucsb.edu; [2]Applied Physics Laboratory (APL), Laurel MD, 20723; [3]Johns Hopkins School of Medicine (SOM), Baltimore, MD, 21205

Introduction

With the current and planned missions to Mars and to some outer planetary moons such as Europa and Titan, NASA is now entering a new phase of planetary exploration strongly motivated by *Astrobiology.* Our interest in Mars is, in part, a result of recent studies of martian meteorites that suggest that the early history of the red planet was remarkably similar to that of the Earth, where life apparently arose both quickly and early on (possibly as soon as ~3.85 Ga). If this is indeed the case, then Mars was presumably a much warmer, wetter, planet then it is today. This hypothesis is further supported by the images returned by the Viking, Mariner and Mars Global Surveyor (MGS) orbiter spacecrafts that all show compelling evidence that copious liquid water existed on the surface of Mars in the past. In fact, new images provided by MGS suggest that there may be current sources of liquid water and ice at or near the surface of the red planet. Other MGS images show evidence of an early ocean at the North Pole and extensive underwater channels draining into large valleys (Valles Marineris) near the equator. All of these data support the possibility that life may have arisen on Mars in liquid water environments.

The search for organic matter in rocks, sediments and ices from Mars is critical to the assessment of any extinct or extant life. Our missions to Mars have the potential to further address the question of whether life arises spontaneously, given appropriate planetary conditions, as well as possibly learning more about our own prebiotic evolution that has been all but erased from the Earth's crustal record. The search for life beyond our own planet is one of considerable interest to scientists and the general public alike. Yet, as we learned from the Viking missions, the search for life signs in a complex environment is problematic and requires an appropriate strategy that will maximize our opportunities to properly examine these compelling questions. This strategy must encompass both the selection of appropriate measurement techniques and the careful testing and evaluation of those techniques in an environment with challenges similar to those found *in situ*.

One of the highest technical barriers to obtaining sensitive analyses of solid phase materials (e.g. ices) on Mars is the often complex and resource-intensive process of sampling. Powerful instruments, such as mass spectrometers, typically require solid samples to be cleanly manipulated and vacuum processed in order to achieve their advertised capabilities. We are in the process of developing and testing an integrated mass spectrometer system that uses a direct sampling and ionization method operating at ambient atmospheric pressure, yet achieves the very high sensitivity and discriminatory power required for complex *in situ* samples. The atmospheric pressure matrix assisted laser desorption/ionization (AP-MALDI) method uses a pulsed laser to volatilize and ionize organic compounds from the surfaces of solid samples, which are then immediately drawn into a differentially-pumped miniature mass spectrometer inlet for analysis.

The development of a MALDI source that operates at ambient atmospheric pressure was strongly motivated by the drive toward higher throughput screening analyses in proteomics and chemical/biological agent detection. In the case of AP-MALDI, the ions are formed at ambient pressure and then drawn into the system. By eliminating the sample acquisition and vacuum loading steps, AP-MALDI has the potential to be used on large sample arrays or wide-area collection plates within a robotic monitoring system or directly on rocks and ice in the field. Moreover, there are a number of features of AP-MALDI that are especially advantageous for use on geological samples in a harsh, remote environment such as Mars. **First,** ambient laser desorption maintains the pristine nature of samples, and the capillary inlet does not contact the sample. Therefore, AP-MALDI could be implemented at the end of an articulated robot arm. **Second,** laser desorption is able to perform local analyses of undisturbed samples that are heterogeneous at the grain scale; the focal diameter is typically in the range from 100 microns to 1 mm. Thus highly localized mineralogical "niches" that may contain biomarker organics are more likely to be detected than with a bulk sample. **Third,** the Mars ambient pressure of 5-10 Torr is precisely in the range that has been shown to dramatically increase sensitivity and decrease metastable fragmentation. That is, a method that provides increased performance at a complexity and convenience cost in terrestrial AP-MALDI may be available without those costs on Mars. The lower ambient pressure (5–10 vs. 760 Torr) at Mars also reduces the overall complexity of the instrument inlet and the requirements on the pumping system because of the reduced differential pressure gradient to reach the required base pressure of the analyzer.

Earth Analogs – Antarctica

Since the exploration of Mars will necessitate a highly robust and refined instrument technology testing equipment in the appropriate Earth analog, like the environments encountered in Antarctica, would greatly facilitate the development of *in situ* instrumentation. Exploration of the ice cap and the unique preservation of organic material in the Antarctic environment will provide the testing ground for the development of our AP- MALDI instrument. Several studies of Antarctic ice have revealed both extraterrestrial and terrestrial sources of organic compounds that are concentrated in the surface ice by the natural process of sublimation (Becker et al., 1999, 2002). The cold and dry Antarctic conditions are ideal for evaluating the preservation of organic compounds and the effects of seasonal changes that may lead to the decomposition of biologically relevant compounds. The acquisition of this data set, both through *in situ* field measurements and with samples collected and returned to the laboratory, will be invaluable to our strategy for the search of extinct/extant organic compounds on Mars.

The current expectation of the ability of AP-MALDI to perform these analyses is based upon initial laboratory work with a breadboard, which has been used to detect 10–50 femtomole (fmol) of analyte (liquid) deposited on a target surface in a four-component mixture of peptides in the 800-1700 Dalton (Da) molecular weight range. Recent analyses of portions of Vostok ice cores revealed between 2 x 10^2 and 3 x 10^2 bacterial cells per milliliter and low concentrations of potential growth nutrients (Karl et al., 1999; Priscu et al., 1999) suggesting that Lake Vostok may contain viable microorganisms. We have available, samples of accreted ice from Lake Vostok. We will present preliminary results on these ice samples and other materials (minerals, sediments) using our AP-MALDI functional proto-type instrument. Testing of the prototype instrument in Antarctica will begin in 2003. This initial testing program will be used to implement the design of our brassboard instrument that will be tested again in Antarctica in the 2006 field season.

References: Becker, L. Popp, B., Rust, T. and Bada, J. L. (1999) *Adv. Space Res.* 24, 477-488. Becker, L. (2002) *National Research Council, Signs of Life: A report based on the April 2000 Life Detection Workshop on Life Detection Techniques,* National Academies Press, Washington, D.C. Karl, D. M., Bird, D. F., Bjorkman, K., Houlihan, T., Shackelford, R., Tupas, L. (1999) *Science* 286, 2144-47. Priscu, J.C., E.E. Adams, W.B. Lyons, M.A. Voytek, D.W. Mogk, R.L. Brown, C.P. McKay, C.D. Takacs, K.A. Welch, C.F. Wolf, J.D. Kirstein. R. Avci. (1999) *Science* 286, 2141-2144.

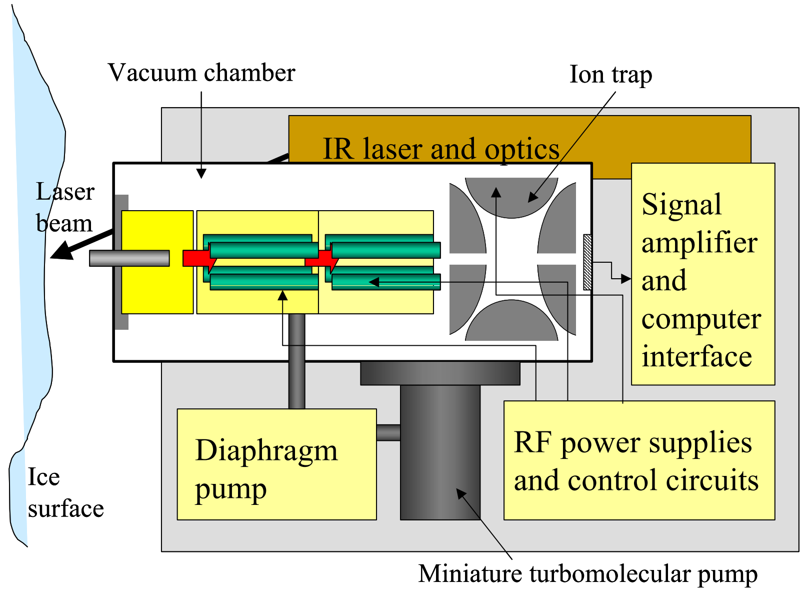

Fig. 1 The envisioned integrated brassboard AP-MALDI-ITMS concept. The instrument analyzes an ice or ice matrix sample deposit in situ, in a non-contact "stand-off" mode. The sample-inlet distance in the mm range depends on a number of factors to be investigated. Laser- sampled ions travel through the inlet stage and focused by a quadrupole/octopole rod assembly to maximize signal. The laser, ion trap, power, and data collection systems are controlled through a user-monitored interface. The asynchronous operation of laser and ion trap permit analyses to be performed in autonomous programmed modes.