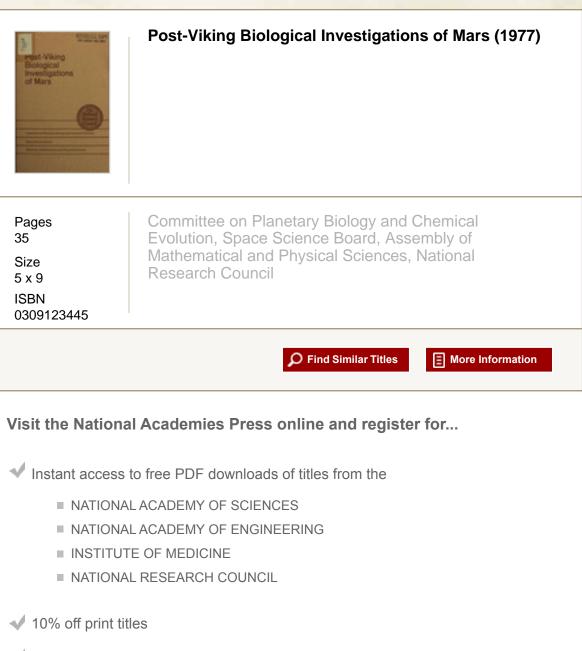
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Post-Viking Biological Investigations of Mars

Committee on Planetary Biology and Chemical Evolution 'Space Science Board 'Assembly of Mathematical and Physical Sciences National Research Council

NATIONAL ACADEMY OF SCIENCES Washington, D.C. 1977

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NOTICE: The project that is the subject of this report was approved by the Governing Board of the National Research Council, whose members are drawn from the Councils of the National Academy of Sciences, the National Academy of Engineering, and the Institute of Medicine. The members of the Committee responsible for the report were chosen for their special competences and with regard for appropriate balance.

This report has been reviewed by a group other than the authors according to procedures approved by a Report Review Committee consisting of members of the National Academy of Sciences, the National Academy of Engineering, and the Institute of Medicine.

FOREWORD

As the Viking mission approached, the Space Science Board become increasingly aware that the problems associated with questions of possible biology and prebiotic chemistry on other planets in the solar system were different from those involved with the use of space to study problems of terrestrial biology and different from the medical and biological problems attending the presence of man in space. Accordingly, in 1973 the Board established an Ad Hoc Exobiology Panel under its Committee of Space Biology and Medicine. In 1975 the Panel became standing, and in 1976 it was constituted as a separate Committee of the Board and renamed the Committee on Planetary Biology and Chemical Evolution.

One major responsibility assigned by the Board to the Panel and then to the Committee has been to monitor the progress of the biologically relevant experiments on Viking, both before launch and after the landing, and to develop a recommended strategy for post-Viking biological investigation of Mars. This report constitutes their evaluation and their recommendations. The report has been reviewed in detail and *in toto* by the Board and was approved unanimously on April 5, 1977. Therefore, both the findings and the recommendations in the report represent the views of the Space Science Board.

> A. G. W. CAMERON, *Chairman* Space Science Board

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PREFACE

The current Viking mission emphasizes the prominent position that has been accorded to Mars in the exploration of our solar system. One reason for this prominence, and for Viking, is that Mars, among all the extraterrestrial objects orbiting the sun, is deemed the most likely to have, or to have had, living inhabitants. The discovery and characterization of present or prior life on Mars would, in the opinion of many, constitute a scientific finding of unparalleled significance to biology, and it would constitute a finding of major importance to planetology, especially to an understanding of the evolution of differences among the planets Venus, Earth, and Mars. For these reasons Viking carried several experiments that were designed to yield information of direct and indirect biological significance. And it is for these reasons that the biological implications of the Viking findings must be weighed in formulating the strategy for the next stages of the exploration of the solar system.

The Space Science Board charged our Committee with evaluating the biological implications of the Viking mission, and it requested us to develop recommendations for post-Viking biological investigations of Mars. This report constitutes both our evaluation and our recommended strategy.

As our report developed, its contents were made available to the Board's Committee on Planetary and Lunar Exploration (COMPLEX), which is currently completing a report of its recommendations on science strategy in general for the inner solar system, including Mars.

About two years ago, the Space Science Board decided that Board reports on strategy for future space science should state the high priority scientific questions that require resolution and the sequence in which they should be investigated and should give some indication as to what measurements need to be made, with what precision, and by what methods. Equally important, the Board decided that the strategy reports should avoid to the maximum extent possible describing strategies in terms of specific flight missions, specific payloads, and specific tactics for missions. Our Committee's report was prepared to be consistent with that policy. As specific missions are conceived and developed by NASA, the strategy documents will be used by the Board (and hopefully by NASA, the Congress, and the public as well) to gauge the scientific importance and appropriateness of the specific missions and their experimental payloads.

The Committee obtained the necessary background for its tasks in a variety of ways. First, it met five times in 1975 and 1976 with members of the Viking Biology Team (H. P. Klein, N. H. Horowitz, J. Lederberg, G. V. Levin, V. I. Oyama, A. Rich, and their colleagues) in Washington, D.C., and at Boston University, the Jet Propulsion Laboratory, and the Ames Research Center. The meetings before the Viking landings were devoted to the nature of the instruments and the design and methods of the biology and organic analysis experiments, to the results obtained when the experiments were conducted on terrestrial soil samples, and to the tactics that the Biology Team expected to apply to the diagnosis of data returned from the surface of Mars. The meetings after the landings were devoted to discussions of the findings from Mars and their interpretation and to discussions of ongoing Earth-based simulation experiments. Second, the Committee attended two and a half days of presentations at the Jet Propulsion Laboratory and the California Institute of Technology on the results of the other Viking experiments. Third, in the fall of 1976, members of the Committee participated in two meetings of COMPLEX, meetings that were concerned with the development of a strategy for the exploration of the entire inner solar system (Mercury, Venus, Earth, Mars, Moon, asteroids, and comets). Finally, we received information and contributions to our understanding of the facts and their interpretation from consultants invited to our meetings and from personal communications with Viking scientists and other colleagues. Specifically, in this regard, we would like to acknowledge the contributions of N. Anderson, K. Biemann, F. Brown, J. Buchanan, S. Chang, F. Cocks, D. DesMarais, D. DeVincenzi, C. Farmer, N. Horowitz, D. Hunten, R. Johnson, H. Kieffer, H. Klein, J. Lawless, J. Martin, E. Merek, T. Owen, R. Setlow, G. Soffen, P. Toulmin, T. Wydeven, and R. Young, as well as members of the Space Science Board.

In addition, the Committee wishes to acknowledge the invaluable role played by its Executive Secretary, Mr. Milton Rosen, and his staff in organizing and guiding our meetings and in the preparation of our report.

This report represents the unanimous view of the Committee. We hope it conveys some sense of the excitement we have felt over the privilege of being accorded an intimate view of the Viking mission, and we hope that it conveys a sense of our enthusiasm for the biological questions that future missions to Mars will surely address over the next decade.

> PETER MAZUR, *Chairman* Committee on Planetary Biology and Chemical Evolution

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INTRODUCTION

The predecessors to this Committee stated in 1974 that "At the present time, Mars is the only real target for exobiological searches in the solar system. All other objects, with the possible exception of Titan, appear to be excluded as possible habitats of life, owing either to the lack of an atmosphere or to temperature regimes that are incompatible with complex organic chemistry. This being the case, the return of unambiguous biological data, either positive or negative, from the two Viking '75 spacecraft can be expected to have a major impact on the planetary program. A positive result will initiate a new scientific discipline, that of Martian biology. A negative result may terminate the search for extraterrestrial life as a motivation for planetary exploration, although interest will remain in the organic chemistry of the solar system." They went on to conclude that "most of the conceivable outcomes of Viking are likely to be viewed as ambiguous or unconvincing," and that "It is difficult to plan responses for this contingency since much depends on the precise nature of the ambiguity."¹, p. 172

Much of the Viking data is now in hand, and we are charged with evaluating its biological significance and making recommendations for post-Viking biological investigations of Mars in the next decade. The evaluation is dealt with in Part I; the strategic considerations in Part II; and our recommendations in Part III.

The discussion will be restricted to Mars. From the point of view of formulating strategy for planetary biology in the next decade, we continue to support the opening sentence in the above quotation that, at the present time, Mars is the only real target for exobiological searches in the solar system. While no one can rigorously exclude the possibility of indigenous life forms on the outer planets, the possibilities are too remote and too little is known about the detailed structure and composition of the planets to justify the direct search for existing life at this time. Nevertheless, an understanding of the organic chemistry of the atmospheres of the outer planets is of major biological interest, and gaining that understanding should remain a high priority goal.

There are three possibilities for Mars: life exists; life evolved but no longer exists; life never evolved. The discovery of existing life would be tremendously exciting. But the other two possibilities would also represent dis-

coveries of profound importance. Venus, Earth, and Mars are roughly similar in size, mass, and distance from the sun. Yet, Venus has a massive atmosphere rich in CO_2 and a surface that is an inferno; Mars has a wisp of an atmosphere (also rich in CO_2) and a surface that is cold, devoid of liquid water, and exposed to highly reactive molecules and intense ultraviolet radiation. Earth has an atmosphere intermediate in density, low in CO_2 , and rich in oxygen and nitrogen. Its surface temperatures lie predominantly in the range where water is liquid, and a great proportion of its surface is covered with liquid water. And most significant of all, it teems with life.

It is customary to think that life exists only on planets that provide the proper conditions for its maintenance. But the realization is growing that life itself may modify a planet's surface and atmosphere to optimize conditions for its existence. Even if it were demonstrated that life does not now exist on Mars, the question would remain whether Earth and Mars differed sufficiently in their early histories to permit the origin of life on the former but not the latter. Or, alternatively, did both planets permit the origin of life and then diverge dramatically? If so, did the type and extent of life that evolved play a major role in that divergence?

These questions are of fundamental scientific interest, but they may also be questions of fundamental importance to all of us on Earth. We have clearly reached the point where human activities are exerting global effects on the composition of the Earth's atmosphere and perhaps its temperature. Atmospheric pollutants may affect the ozone layer and could modify the Earth's albedo. The burning of fossil fuels has already measurably increased the carbon dioxide content of the atmosphere, and some scenarios predict serious and even devastating consequences if major fractions of our energy requirements continue to be derived from these sources.² Clearly the stability of equilibria and steady-state processes on the Earth's surface and in its atmosphere to human perturbants, and the role of the Earth's biota in this stability are matters of more than arcane interest. Since the surface of Mars provides a natural global system for comparison with Earth, we submit that studies of biology and of chemical evolution on our neighboring planet will shed important light on these terrestrial questions-questions that could be significant to our ultimate survival.

I SUMMARY OF VIKING FINDINGS RELEVANT TO MARTIAN BIOLOGY

The results of several of the Viking experiments are relevant to questions of current or past life on Mars. We summarize in this section information that seems especially pertinent.

A. Elements

The discovery of nitrogen in the atmosphere has eliminated a major barrier to the postulation of the existence of Martian biota. There is general agreement that the absence of nitrogen would have constituted a definitive global negative for current life. Calcium, sulfur, magnesium, chlorine, and probably potassium and phosphorus have also been detected in soil* samples.³ All six (and especially phosphorus) are likely to be essential to living systems.

B. Water (General)

1. Major geographic, topographic, and diurnal variations in the concentrations of atmospheric water vapor have been observed, reaching values of 75 precipitable micrometers or higher near the North Pole. The relative humidity at the surface is unknown except in the North Polar region where orbital observations and calculations indicate saturation.⁴

2. The residual North Polar cap has been shown to be water ice, probably 1 to 1000 m or more thick.^{4,5}

3. Several experiments confirm or strengthen the inference that large amounts of water are locked beneath the surface in the form of a permafrost. Analysis of the composition of the atmosphere suggests that the total volatile inventory on Mars may be much larger than the content of the present atmosphere. Two independent lines of argument have been proposed.⁶ One follows from the observed enrichment relative to terrestrial and solar abundances of ¹⁵ N to ¹⁴ N. If this enrichment has resulted chiefly from preferential escape of the lighter isotope from the upper atmosphere, it indicates that

*Because the word "soil" implies the presence of organic compounds and connotes a material that will support terrestrial plant life, we will chiefly use the more neutral term "regolith," which is defined as unconsolidated planetary surface material, i.e., rocky rubble.

Mars must have had at least 10 times the present abundance of N_2 . This same escape process should also have led to an enrichment of ¹⁸O, which is not observed. To prevent the enrichment from occurring, one needs a large reservoir of oxygen, and the most likely reservoir would be water in amounts equivalent to at least 2 bars of vapor. Similar estimates of the size of the water reservoir are arrived at if one assumes that water and nitrogen outgassed simultaneously.

A second proposed line of argument⁶ follows from the detection of ³⁶Ar, Kr, and Xe in the Martian atmosphere. These gases demonstrate that Mars has outgassed by a factor 100 times less than on Earth. Comparison of the ratios of ³⁶Ar/N₂/CO₂/H₂O on Earth and Mars indicates that the present Martian atmosphere is deficient in N₂, CO₂, and H₂O. Obtaining a match would require that the planet once had at least 10 times more CO₂ and N₂ than is now seen in the atmosphere.

Both lines of evidence suggest Mars once had water equivalent to a layer of 20-30 m over the entire planet. Amounts equivalent up to perhaps 2 m of global coverage are locked up in the residual polar caps; amounts equivalent to perhaps 2 m have probably photolyzed and escaped from the planet. The rest must be buried in the regolith.⁶

4. The diurnal behavior of water vapor in the atmosphere indicates that most of it is located near the surface and that at least 80 percent of the vapor returns to the solid phase between noon and the following dawn. The rate of reappearance of the vapor at dawn is sufficiently slow to require that this solid phase be beneath the surface.⁷ The residual North Polar cap appears to represent a region where the permafrost "breaks through" to the surface.⁴

5. The gas chromatograph-mass spectrometer (GCMS) on Viking has detected 0.1 to 1.0 percent (w/w) water in regolith samples heated to 350° C or 500° C, but with one exception much less than 0.1 percent water in samples heated to 200° C.⁸ The results are consistent with the water being that in mineral hydrates of moderate thermal stability (perhaps hydrates of MgSO₄³). The one exception is the sample collected by the second lander (VL-2) from beneath a rock. It yielded no water when heated to 50° C but several tenths of a percent when heated to 200° C. It could represent tightly adsorbed water or mineral hydrate water of low thermal stability.

C. Water (Liquid)

1. Past liquid water. Evidence from orbital photographs is powerful that massive quantities of liquid water once existed and flowed on the Martian surface. Flowing liquid water means the existence of sediments. However, estimates from crater counts suggest that the channels were formed a billion or more years ago.⁹ If so, the relevance of the fluvial areas to the existence

of *current* life is dubious, although their relevance to the possible existence of past life and to organic chemical evolution may be profound. The topographical evidence for surface liquid water in the past is consistent with the conclusions derived from analyses of isotopic ratios of atmospheric nitrogen and argon. These analyses indicate that the total surface pressures on Mars could easily have been high enough to have permitted the existence of surface liquid water.

2. Present liquid water. Liquid water is generally agreed to be essential for the functioning of living forms. But, unfortunately, Viking was not designed to detect free liquid water and has not done so. Liquid water adsorbed to the soil ought to have been detectable in the 200°C pyrolysis in the GCMS, but less than 0.1 percent was detected (several tenths percent in the subrock sample), and that could represent mineral hydrate water of moderate or low thermal stability.⁸

The detailed measurements of surface temperatures and atmospheric pressures continue to preclude the existence of pure bulk water under equilibrium conditions. The three possibilities for liquid water proposed prior to Viking still remain remote possibilities: (1) liquid water adsorbed to subsoil; (2) water that is liquid by virtue of kinetic factors slowing the approach to equilibrium (i.e., conditions under which diffusion of water is slower than diffusion of heat¹⁰); (3) water that has its chemical potential (and hence freezing point) lowered by the presence of dissolved solutes. Possibility (3) has been enhanced by the X-ray fluorescence detection of elements like Ca, Mg, Cl, and S that could give rise to water-soluble ions.³ (In fact the existence of MgSO₄ at the two landing sites is now considered likely.) Salts like CaCl₂, MgCl₂, and K₂CO₃ have eutectic points below -30° C, and their presence would permit stable liquid water down to these temperatures. The electrolyte concentrations, however, would be multimolar.

Even though data from the Viking landers have neither lessened nor especially enhanced the possibility of stable or metastable liquid water in the regolith, the surface and subsurface temperatures that have been estimated from orbital infrared measurements, in conjunction with the low atmospheric pressures, continue to make that likelihood remote. Summer surface temperatures at the landing sites vary diurnally between -88° C and -3° C. Some 24 cm below the surface the summer temperatures are expected to be a steadier -51° C to -56° C. In the winter, the surface temperatures at VL-1 are expected to vary diurnally between -95° C and -22° C, and those at VL-2 between -124° C and -82° C. Some 24 cm below the surface the winter temperatures at VL-1 and VL-2 will be -69° C and -108° C, respectively.¹¹ Mechanisms for providing liquid water below -50° C become increasingly limited; no mechanisms are known to provide liquid water below about -70° C. To give some feeling as to the harshness of these temperatures to terrestrial biota, the

lowest confirmed minimum growth temperature for Earth organisms is -15° C, but very few can grow below 0°C. Interlamellar layers of water adsorbed on soils remain unfrozen to at least -30° C,¹² but the very forces that keep the water unfrozen make it a difficult source for organisms.

D. Reduced Carbon and Organics

1. With the possible exception of data from the pyrolytic release experiment, there continues to be no evidence for the existence of carbon reduced below the state of CO and no clear evidence of any form of carbon save in the atmosphere and in the winter polar caps.

2. Organic compounds. No organic compounds, other than traces attributable to terrestrial contaminants, have been detected in regolith samples analyzed by the GCMS. If volatizable organic compounds were present in the samples, they were either present in concentrations below the parts per billion range (the detection limit of the instrument) or they were totally restricted to substances like methane with molecular weights of less than 18. which are undetectable or detectable only at reduced sensitivities. (A third possibility, the complete oxidation of organics during heating in the sample chambers, is considered by the molecular analysis team to be very unlikely.⁸ One argument presented is that known terrestrial organic contaminants, methyl chloride, acetone, toluene, and benzene, were detected in expected amounts during the experimental runs on the Martian samples.) Instruments with the same characteristics as the flight instrument have invariably detected organic compounds in all terrestrial soil samples tested, including antarctic soils with few living organisms. The concentrations of organics, if present in Martian samples, appear less than the concentration of organics in lunar samples.¹³ The concentrations are less than those expected from the influx of carbonaceous chondrites (assuming the regolith is mixed to a depth of 100 m or less⁸). This latter conclusion, combined with two lines of evidence for the existence of strong oxidants in the regolith, leads to the view, that in at least the top few centimeters of the surface, carbon-carbon bonds are disrupted faster than they are deposited or synthesized. The first line of evidence comes from orbital measurements of the atmosphere and modeling. One model predicts the existence of active strongly oxidizing species, especially hydrogen peroxide.¹⁴ Second, the gas exchange experiment (GEX) on the Viking landers showed the release of up to nearly a micromole of oxygen when the \sim 1-cc soil samples were humidified with water and warmed to $\sim 10^{\circ} C^{15,15a}$ (see below).

The inability to detect organic compounds in the regolith samples does not itself exclude the possible existence of a microbial population. Fewer than 10^5 to 10^6 representative terrestrial bacterial cells would not contain sufficient organics to be detectable with the GCMS.⁸ However, postulating the

existence of a viable microbial population under such conditions requires speculative scenarios. On Earth the great bulk of the organic compounds in soils represents the transformed remnants and metabolic products of the organisms and not the organic content of the living organisms themselves. Thus, the ratio of organic carbon in living microorganisms to that in humus is about 1 percent, and the ratio of organic carbon in living microorganisms to the total organic and elemental carbon in oil, gas, coal, oil shale, humus, and in the oceans is estimated to be 0.0001 percent to 0.001 percent.^{2,16}

If Martian surface samples in fact contain living microbes, one must assume that mechanisms exist which permit their existence, while at the same time preventing the buildup of their organic detritus to levels detectable in the GCMS. There are possibilities such as (a) the transport of living organisms from other, more hospitable areas at a rate sufficient to balance the destructive processes, and thereby provide a steady-state population of viable cells; (b) efficient recycling of organic detritus by the microbial population; or (c) the organisms possess biologically driven devices or mechanisms to protect their organic matter, and these devices and mechanisms disappear upon their death. The best that can be said for items (a) and (c) is that there are partial Earth analogues. However, on Earth we know that hospitable areas exist; and on Earth samples from even harsh environments contain detectable organic compounds, and they possess ratios of total organics to the organics in living cells that far exceed unity. Possibility (b) is not especially helpful unless the recycling efficiencies approach 100 percent.

E. Biology Experiments

The biology package in each Viking lander contains three separate experiments: gas exchange (GEX), labeled release (LR), and pyrolytic release (PR). The first two provide the Martian samples with water vapor at high activity or with liquid water containing organic substrates commonly used by terrestrial microorganisms. The third provides only two gases known to be constituents of the Martian atmosphere (CO and CO₂), light (optional), and small amounts of water vapor (optional).* In all three experiments, the samples have so far been incubated at 8°C to 26°C. The significant measurements are the quantity of gas(es) evolved (GEX, LR, and PR), the type of gas (GEX), or

*In the GEX experiment, the amount of aqueous nutrient medium (0.6 cm^3) added initially to the bottom of the test cell is such that the regolith sample is contacted by water vapor only and not by the liquid medium. Subsequent additions of medium actually wet the sample. In the LR experiment, 0.115 ml of liquid medium is added initially to 0.5 cm³ of sample, and the liquid contacts only the central core of the sample. Subsequent additions of medium wet the entire sample. The PR experiment is run either without the addition of any water or with the addition of about 80 μg of water vapor to 0.25 cm³ of soil in the 4 cm³ test cell.

the kinetics of its evolution (GEX and LR). In PR the samples are heated in such a way after incubation and the volatiles passed through a trap of such characteristics that the detection of $^{14}CO_2$ in the so-called "second peak" is presumptive evidence for the synthesis of organic compounds during the incubation of the sample. The experiment is designed to test the samples "for the presence of microorganisms by measuring the incorporation of radioactive CO_2 and CO into the organic fraction of a soil sample."

In GEX and LR the evolution of gases indicates that reactants in the samples or added reactants have undergone chemical reactions. The assumption is that microbial activity could be diagnosed from the amounts, or types, of evolved gases and from the kinetics of their appearance.

All three experiments have yielded signals that clearly indicate chemical activity.¹⁵ What is less clear is the interpretation of the signals. Some aspects of the data are consistent with those expected from biological activity comparable to that observed on Earth, but other aspects are inconsistent. In the LR experiment, the production of ¹⁴ CO₂ when regolith samples were initially moistened with a nutrient medium is consistent with biological activity.¹⁷ So also is the synthesis in the PR experiment of picomole quantities of organic matter during the 120-h incubation of samples in the light.¹⁵ (However, although statistically significant, the amount synthesized in the PR experiment is only about one-tenth that synthesized by terrestrial soils that give the minimal observed response, i.e., antarctic soils.^{18,19}) In both the LR and PR experiments, the activity was abolished or appreciably reduced when the regolith samples were preheated to 170°C to 180°C for 3 h. Heating to such temperatures abolishes biological activity in terrestrial samples.

A major difficulty with a biological interpretation of the data is the response of samples to water. Rather than enhancing the signal as expected for biological activity, the addition of water vapor in the PR experiment totally prevented the reaction from occurring.¹⁸* In the LR experiment, the initial

*This conclusion has now been contradicted by the results of the final PR experiment, results received after completion of this report. The last three runs (C-4, C-5, and C-6) were performed on aliquots of a sample collected by the VL-1 lander and stored at 5°C to 24°C for 0, 71, and 143 (Earth) days, respectively. All three gave statistically identical values for the "second peak," in spite of wide differences in their thermal and water treatment. Sample C-4 was run dry at 16°C in the usual fashion; sample C-5 received water vapor, and was then vented, heated to 90°C for 112 min, and finally incubated at 17°C; sample C-6 received water vapor, and was then incubated at 15°C. The response of this last sample differed dramatically from the results of the previous sample (U-2) run in the presence of water, a run that had given a second peak of 0. The discrepancy remains unexplained. Horowitz, Hobby, and Hubbard (personal communication, 1977) conclude, however, that "a biological interpretation of the [PR] results is unlikely in view of the thermostability of the reaction." Even after a sample was heated to 175°C for 3 h, it yielded a second peak count that was 12 to 60 percent of that in unheated samples. addition of medium, which wet only a portion of the sample (footnote, page 7), appeared to exhaust the reactants in the entire sample.^{15,17} Further inconsistencies emerge from the results of the GEX. It showed the unexpected release of as much as 0.7 μ moles of molecular oxygen, indicating the presence of strong oxidants in the samples (probably peroxides or superoxides—see below). Here too the initial introduction of water vapor exhausted the reactants that were the source of the oxygen, i.e., further additions of liquid aqueous medium produced no further evolution of oxygen.^{15,15a}

Intensive attempts are now being made to simulate the results of the Viking biology experiments abiologically. Although the information is preliminary, major features of the LR and GEX experiments have been mimicked at least qualitatively by nonbiological reactions. The major feature of the LR experimental results is the decarboxylation of the organic substrates to yield CO_2 . A number of strong oxidants like hydrogen peroxide and metal peroxides and superoxides are known to drive that reaction.^{19a,20} Carbon dioxide is also evolved when either formate or the nutrient mixture used in the LR experiment is subjected to intense UV radiation in the presence of ferric oxide.²⁰

The major feature of the GEX experimental results is the release of oxygen when the Martian samples are humidified. Once again, a number of metal peroxides and superoxides evolve oxygen when placed in contact with water.^{20, 20a} Wydeven and his colleagues^{20a} have also obtained oxygen evolution in amounts and at a rate comparable to that observed in GEX on Mars by exposing soil samples in a GEX experiment on Earth to a gas mixture obtained by passage of oxygen through a radio frequency glow discharge. The RF treatment produces active species of oxygen similar to those expected to be generated at the Martian surface by solar UV radiation. The latter process has been modeled in some detail.¹⁴ Splitting of H₂O gives H and OH, which leads by well-characterized pathways to the production of H₂O₂.

To date, the results of the PR experiment have not been simulated abiologically, and possible abiological explanations are speculative. It seems clear that picomole quantities of organic compounds were synthesized in several of the PR experiments. The specific observations were that after the Martian samples (no H₂O added) were irradiated at \geq 320 nm (0.5 percent <320) in the presence of ¹⁴CO₂ and ¹⁴CO, and then pyrolyzed at 625°C, significant quantities of ¹⁴C-containing material were retained on the organic vapor trap (OVT) at 120°C. Heating of the OVT to 650°C released this material and oxidized it to ¹⁴CO₂ where it was detected as the "2nd" peak. Experiments with terrestrial soils have shown: (1) the ability of the OVT to retain organics other than gaseous forms like methane or ethane; (2) high efficiency of the OVT in passing through unreacted ¹⁴CO₂ and ¹⁴CO (less than 0.01 percent is retained); (3) the PR experiment yields positive results in all terrestrial soils

shown to contain viable cells (provided that water is present); and (4) it yields negative results in sterilized soils.^{19,21}

It is known, however, from the work of Hubbard *et al.*²¹ that nonbiological organic synthesis can occur under conditions analogous in several respects to those that prevailed in the PR experiment. They have found that, in the presence of solids of high surface area, formic acid and other organic compounds are synthesized when CO, CO₂, and small amounts of H₂O vapor are irradiated at wavelengths of 250-280 nm. Very little abiogenic synthesis occurs at longer wavelengths, and it was for this reason that only wavelengths longer than 320 nm were allowed to reach the samples in the Viking PR experiment. But perhaps on Mars, these longer wavelengths can drive the reaction. For instance, as mentioned, hydrogen peroxide is likely to be present in the Martian regolith.¹⁴ Hydrogen peroxide dissociates into hydroxyl radicals when irradiated with even visible light (quantum yield 0.3 at 313 nm and measurable reactivity at 365 nm²³). And hydroxyl radicals have been implicated²² in the abiogenic syntheses observed by Hubbard *et al.*

Possibly then the reaction occurring in the PR experiment on Mars is something akin to $H_2O_2 + CO^{hy}$ formic acid. (This illustrative reaction is at least thermodynamically feasible, for it has a free energy of -20 Kcal/mol.²⁴) Reacting along this pathway, however, would require special conditions, for the oxidative pathway, which yields CO_2 and H_2O , has a free energy of -90 Kcal/mol.²⁴

We are not proposing that this is necessarily the actual reaction that occurred in the PR experiment. We cite it to illustrate two points. One point is that abiogenic explanations of the PR results are conceivable. The second point (applicable to LR and GEX as well) is to emphasize that conditions at the regolith-atmosphere interface on Mars are vastly different from those at the soil-atmosphere interface on Earth. This vast and incompletely characterized difference makes it inordinately difficult to conclude that experimental results, which are unambiguously ascribable to biological activity on Earth, are unambiguously ascribable to biological activity on Mars. The converse is also true. The incompletely characterized differences will make it difficult to determine rigorously whether the Viking biology results have been generated abiologically. The ambiguities are likely to remain despite continued experimentation on the Viking landers and despite further efforts at terrestrial simulation. Nevertheless, we consider further efforts on simulation to be vital, for they will permit the ambiguities to be reduced to a minimum.

Despite current (and possibly future) inability to reach rigorous interpretations of the results of the Viking biology experiments, our Committee is charged with recommending strategy for the future biological exploration of Mars. Our judgment is that the evidence at hand is sufficiently persuasive

to require that that strategy be predicated on the assumption that the positive signals from Mars are not biological in origin. That judgment rests chiefly on six points: (1) the evidence from GEX for the presence of strong oxidants; (2) the inhibitory or dissipative effects of the presence of added water (but see footnote, p. 8); (3) the lack of detected organic compounds; (4) the ability to account, at least qualitatively, for the results of GEX and LR by nonbiological reactions; (5) the prior demonstration that abiological organic synthesis can occur under conditions analogous to those in the PR experiment except for the wavelength of the incident radiation; and (6) the existence of at least conceivable mechanisms for a different wavelength dependence at the Martian surface.

We wish to emphasize that we cannot draw conclusions as to whether life currently does/or does not exist on Mars. Although increasingly unlikely, the positive signals from one or more of the experiments *could* be biological in origin; a second possibility (also remote in our view) is that, although the signals are abiogenic, life in fact exists at the landing sites but was undetected. A third possibility is that, although the samples are lifeless, life exists elsewhere on the planet's surface or beneath its surface. A fourth possibility is that life evolved but no longer exists. Finally, the fifth possibility is that it never evolved. As mentioned in the introduction, the last three possibilities are all questions of fundamental biological importance, and it is they that form the basis of the ensuing discussion of post-Viking strategies.

II POST-VIKING BIOLOGY STRATEGY FOR MARS

Although Viking has not determined whether life exists on Mars or whether it once existed, the detection of atmospheric nitrogen prevents one from excluding the former possibility. Some of the findings enhance the possibility of current or past life: ancient water flows, the existence of salts, and the near certainty that large amounts of ice are locked in the regolith. Some factors diminish the possibility of current life: the lack of detectable organic compounds or reduced carbon, the presence of strong oxidants in the soil, and the low probability for the existence of liquid water under equilibrium conditions.

A. Criteria for Sample Selection and Characterization

Any biological experiment begins with the selection of samples. If the top few meters of the Martian surface were homogenous over the entire planet, or if variations were randomly distributed, the optimum strategy for the selection of samples would be fairly obvious. But photographs from orbit and from the landing sites have shown that the surface is heterogeneous and that the heterogeneities are not randomly distributed. Furthermore, data from various experiments strongly suggest that the physical and chemical characteristics of the regolith are not uniform with depth.

Inextricably entwined with the question of where to sample is the question of what characteristics of the sample would constitute items of paramount importance to present or past biology and to organic chemical evolution.

We submit that the following fall in this category:

1. Does the sample contain detectable organic compounds or reduced carbon? The distribution, state, and abundance of carbon is critical to the possible origin and current existence of Martian life. Carbon is the fourth element in cosmic abundance. Diverse organic compounds of considerable complexity are distributed throughout our galaxy. It is remarkable, then, that on Mars no carbon has been detected definitively except in the atmosphere and in the winter polar caps, and none has been detected in a form more reduced than CO. The detection of reduced carbon would not prove current or past life (since it could be deposited by carbonaceous chondrites

or it could be synthesized abiogenically), nor, as already discussed, could the lack of detection disprove it. Nevertheless, since reduced carbon is considered a *sine qua non* of living systems, samples with detectable elemental carbon or organic compounds clearly have an enhanced probability of containing past or current life. Characterization of the type of compounds and the determination of ${}^{12}C/{}^{13}C$ ratios in carbonate and in organic matter and the ${}^{32}S/{}^{34}S$ ratios of reduced sulfur and sulfate in minerals 25 might permit some discrimination between biological and nonbiological genesis.

2. Does or could the sample contain liquid water at high chemical potential?* Liquid water is also generally considered to be an absolute prerequisite of living systems. The properties of liquid water are unique and play a major role in determining the conformation and therefore the function of terrestrial macromolecules. Water vapor does not have these unique properties. Ice has many of the properties of liquid water, but its very high viscosity would greatly restrict biochemical reactions, and at sufficiently low temperatures would preclude them.

3. Does the sample contain water-soluble electrolytes? So universal is the presence of electrolytes in terrestrial biological systems and so important is their role in both macromolecular conformation and enzymatic and physiological function, that we consider them a likely essential of all living systems. Especially significant in samples would be ions of Na, K, Mg, Ca, Cl, S, and P. Furthermore, their presence in appreciable concentrations in a sample would enhance possibilities for the existence of liquid water.*

4. Is the composition of the gases in the soil sample out of equilibrium with that in the atmosphere? Biological activity modifies the composition of the surrounding gases. The detection of disequilibria between the bulk atmosphere and the occluded gases in a soil sample would not prove the presence of life, but soil samples exhibiting prolonged disequilibria of changing magnitude would certainly be deemed more likely to contain active organisms. This would be especially so if the gases were those that on Earth cycle biologically critical elements: hydrogen, methane, ammonia, hydrogen sulfide, oxygen, nitrogen, carbon dioxide, nitrogen oxides, and volatile amines.

*Although water retains many liquid properties, such as molecular rotational mobility down to activities as low as perhaps 0.2, nearly all fully functioning terrestrial life requires the chemical potential of liquid water, expressed as activity, to be >0.9. The extreme lower limit is about 0.61 for one species of mold, and there are scattered reports of growth at $a_w < 0.8$. [Water activity = P/P_o , where P is the vapor pressure of the water under consideration, and P_o is the vapor pressure of pure bulk water.]

The presence of solutes reduces the water activity and consequently the freezing point. For example, solute concentrations that lower the water activity to 0.9, the minimum value for the functioning of most terrestrial organisms, also reduce the freezing point of the aqueous solution to about -11° C.

B. Where to Sample

1. Sediments. Clearly Mars once had massive quantities of flowing surface liquid water. Flowing liquid water produces sediments, and it is in these sediments that one would expect a higher probability of the evidence for past life, a higher probability of biologically derived organic compounds, and a higher probability of appreciable concentrations of electrolytes. Obvious candidates for sampling are areas exhibiting past fluvial activity and areas exhibiting sequential layering, such as the margin of the north residual polar cap.

2. Ice-regolith interfaces. It is at such interfaces that the existence of liquid water would have the highest likelihood. One such promising area is again the margin of the residual polar cap, especially perhaps the margins of those frost-free patches in the residual cap that have temperatures as high as 235° K.⁵ Other potential locations for liquid water are the regions lying between the surface and the subsurface permafrost.

3. Subsurface sampling. As of April 1977, Viking has conducted organic analyses and biology experiments only on samples from the top 4-6 cm of the regolith. (In March 1977, an inorganic analysis was carried out on a sample from a depth of 20 cm.) Some of the sampled material had been exposed to Mars' intense ultraviolet flux, and all the material presumably contained strong oxidants derived from atmospheric processes—strong oxidants that would probably have destroyed organic compounds if they had been deposited or synthesized. Clearly, sampling at greater subsurface depths is required to reduce or eliminate the powerful oxidants and thereby enhance the probability of locating organic compounds. Equally clearly, subsurface exploration will likely be required to reach the putative ice-regolith interfaces and sediments just discussed.

The chief argument against subsurface sites for living forms has been that the sites would be reached by little or no visible light and, hence, would be incapable of supporting photosynthetic autotrophy. On the other hand, if the results of the current pyrolytic release experiment represent abiogenic organic synthesis, there may be sufficient steady-state quantities of organic compounds below the Martian surface to support heterotrophy or chemical autotrophy.

In summary, then, from the biological viewpoint, the prime concern for the next mission to Mars should be exploration of the subsurface in favorable areas containing sediments or layered sequences such as alluvial flows, the margins of polar ice caps, and terrain likely to overlay permafrost. Samples collected from these areas should be characterized with respect to the following first-order requisites for current or past living systems: (a) the existence and identification of specific organic compounds; (b) the distribution of ice and liquid water and of water-soluble electrolytes; (c) examination of

the geochemistry and morphology of sedimentary materials; (d) measurements of occluded gases to determine those that are significantly out of equilibrium with the atmosphere; and (e) measurements of the isotopic ratios of carbon in the reduced form, if present, and in carbonate, and measurements of the oxidation state and isotopic ratios of sulfur.

C. Instrumental Requirements for Sample Characterization and Considerations of the Strategy for Search and Sampling

1. Instruments. Instruments of the requisite type, sensitivity, and resolution either have flown on the current Viking or appear adaptable to future soft landers without difficulty. A critical requirement for any instrument is that the data it furnishes be directly interpretable and subject to minimal ambiguity. We cite examples for the purposes of illustration—not to represent definitive recommendations.

(a) Organic compounds, isotope ratios, and gas analysis. Modifications of the existing Viking GCMS would provide an instrument meeting the basic requirements for all three analyses. We are informed that an instrument could be provided with both broad mass number coverage to aid in the identification of organic compounds and on command high resolution and accurate peak height analyses over a restricted mass range, as would be desirable for isotopic and gas analyses. The instrument should be capable of carrying out a large number of analyses, and its detection threshold should be at least equal to that in the present GCMS. Alternative, although perhaps somewhat less versatile, approaches would be gas chromatographs or mass spectrometers by themselves. The state of art for both is high. Both have flown on Viking,^{8, 15} and a second-generation instrument incorporating a mass spectrometer is currently under development.²⁶⁻²⁸

(b) Detection of the amounts and phases of water. A candidate instrument has been proposed and tested in the bread board state. It consists of a differential scanning calorimeter (DSC) coupled with a phosphorus-pentoxide-conductivity water detector. Both components are commercially available. Adaptation to flight configuration is deemed feasible.²⁶ Commercially available instruments can detect the heat absorbed in the melting of $\sim 1 \times 10^{-7}$ g of water. An improvement of two orders of magnitude is probably attainable.²⁹

(c) *Electrolytes.* The presence of electrolytes could be easily detected by suspending regolith samples in water and measuring the electrical conductivity. Such measurements combined with inorganic analyses of the sort conducted on the current Viking would provide information as to the elemental species involved. Also essential would be at least an approximate measure of pH (± 0.5 pH).

(d) Identification and characterization of sediments in fluvial areas. Samples will need to be examined and characterized with respect to determinable major features. One feature would be particle size within horizontal laminae or beds. Another would be the size and shape of the rock or mineral grains. The angularity of fragments would be suggestive of the degree of transport to which the material had been subjected and might allow some characterization of mineral composition or source rock. The imaging systems required to conduct these examinations are considered technically feasible.²⁶

2. Mobility and sampling in depth. We stress the importance of obtaining samples from sedimentary areas, from ice-regolith interfaces, and from areas overlaying permafrost. To locate these and to escape the UV-oxidant conditions at the surface require subsurface sampling. However, the problem of specifying an optimum strategy for lander delivery, search, and sample acquisition is formidable.

(a) Delivery. Potentially interesting sites will be selected from orbital photography and other orbital measurements. But sites of maximum biological interest are among the more hazardous with respect to soft landings. One solution to this quandary is to provide the lander with mobility. But how much mobility? The requirements of safety and the size of the landing error ellipse probably dictate mobilities of many kilometers. But the operational feasibility of achieving such mobilities is moot. The alternative route is hard landers. The penalties here are the restrictions on size, weight, and complexity of the instrumental payload (see below).

(b) Search. A tremendous gap exists between the topographical features that are resolved and interpretable at orbital altitudes and the surface features that are actually found by landers. It is quite conceivable that a site chosen from orbit for highly interesting topographical features such as fluvial activity would upon landing appear indistinguishable from the VL-1 and VL-2 sites, both at the landing site and for a hundred kilometers around. Perhaps candidate sites for sampling will be made self-evident by the visual observation of geological features such as a sedimentary outcropping, but more probably they will not. The likelihood, then, is that specific subsurface sampling sites will have to be chosen stochastically.

(c) Subsurface sampling. Similar problems arise with respect to defining the depths from which samples should be acquired. A few millimeters of regolith would greatly attenuate the UV flux if the regolith were static.^{29a} It is not static, but to our knowledge neither the time scale nor the depth of mixing has been estimated. There are even more serious obstacles to estimating the depths required to reduce or eliminate the powerful surface oxidants, since they may well consist in part of diffusible compounds such as hydrogen peroxide. Finally, we do not at present know whether the subsurface sedi-

ments or regolith-ice interfaces that we wish to locate lie 6 cm or 60 m below the surface.

The technical problems and attendant costs of subsurface sampling by soft landers are formidable, and both are exponential functions of the specified depth. It is essential, then, that NASA conduct studies to estimate or to devise approaches to estimating the required subsurface depths. Perhaps estimates can be derived from data from the current Viking. Perhaps information on the distance to the subsurface ice could be gained from radar or other observations from Earth. Or perhaps the only approach to obtaining useful estimates will be in situ measurements on the Martian surface of regolith properties such as the frequency dependence of conductivity.

The last possibility would lead to difficult choices: Should the exploratory phase which acquires and characterizes subsurface samples for biologically relevant properties be preceded by a mission that is designed in part to estimate such physical attributes as the distance to the ice-regolith interface? Or should the biological exploratory phase be initiated without this prior information? If initiated without the prior information, should the delivery mode be penetrators which could sample at uncontrollable depths down to several meters, or should the delivery mode be a mobile soft lander provided at major additional cost with the ability to sample at controllable sites to controllable depths of perhaps several meters?

3. Penetrators and other hard landers. As mentioned, hard landers offer one approach to sampling in-depth areas that are topographically interesting but too hazardous for soft landers. One type of hard lander, the penetrator, has been evaluated in considerable detail and offers promise.³⁰ The penetrator is a missile-shaped object that impacts the surface at high speed and penetrates to depths of 1 m to 15 m, leaving an afterbody on the surface. The forebody contains most of the scientific payload; the afterbody contains a transmitter and, if desired, an imaging system. A "nominal" penetrator could carry a payload weighing 7 kg and occupying 4500 cm³. The payload is subjected to a deceleration of about $1000 \times g$. The payload capacity (along with limitation in power) puts severe constraints on instrumentation, but the deceleration forces do not. (One preliminary study has indicated that a Viking-type mission could carry perhaps nine such penetrators in lieu of a soft lander. However, the two types of landers are not mutually exclusive, even on a Viking-type mission. The types and numbers of landers transported depend, of course, on the propulsion systems available.)

In spite of the payload restrictions, the instrumentation on penetrators can be remarkably sophisticated. With respect to measurements critical to biology, it could probably carry (1) a DSC-P₂O₅ instrument for detecting amounts and phases of water, (2) apparatus for measuring soil electrical conductivity, and

(3) various instruments to conduct inorganic and elemental analyses. A critical but still moot question is what could be carried in the way of organic analytical instrumentation. The adaptation of a GCMS or a mass spectrometer itself seems unlikely. However, the adaptation of a gas chromatograph seems more feasible. Gas chromatographs can be small, and they are extremely sensitive and capable of good resolution, provided a volatile phase can be generated. Finally, there exist sensitive techniques capable of giving yes or no answers about the presence of organic compounds; for example, spectro-acoustical techniques capable of detecting C-H bonds in picomole quantities now exist,³¹ and they can possibly be adapted to penetrators.

In summary, penetrators have the advantages that they can be directed to numerous areas that are deemed of high biological promise^{*} and that they would provide subsurface sampling. They have the disadvantages that only the impacted sites are sampled, that the vertical range of subsurface sampling is restricted, that the instrumentation is far more restricted than on a soft lander, and that there is far less potential for adaptivity in the conduct of the experiments. The major unknown is their specific instrumental capability. Specific flight-configuration instruments for collecting biologically critical data do not yet exist.

D. Further Considerations Concerning the Strategy of Exploration

To repeat, from the biological viewpoint, the first phase of post-Viking exploration of Mars should be to acquire subsurface samples from areas likely to contain sediments and ice-regolith interfaces, and to characterize these samples with respect to organic content and carbon and sulfur isotope ratios, the abundance and state of water and water-soluble electrolytes, the abundance and types of occluded gases, and the geochemistry and morphology of the sediments. It is not clear whether the necessary characterization can be achieved by a hard lander or a soft lander alone, or whether it will require both, probably in sequence. The reason it is not clear is that there are at least two major areas of ignorance: (1) A soft lander can unquestionably carry the necessary instrumentation to characterize subsurface samples, but there are serious questions as to whether it can reach the areas that are prime candidates for sampling; (2) hard landers can probably reach the areas that are prime candidates for sampling, but there are unresolved questions as to whether

*It should be noted that the error ellipse for the point of impact of penetrators is comparable in size to that for soft landers; hence, penetrators should not be considered capable of hitting small targets. Moreover, although penetrators can carry the variety of instruments listed above, limitations in space and demands from other experiments make it unlikely that each could carry the full complement of biological instruments. they can carry the instrumentation necessary to characterize the samples satisfactorily.

It is our strong recommendation that NASA carefully address the matter of mobility of soft landers and the matter of instrument payload on hard landers before deciding on subsequent missions to the surface of Mars.* Attention has been devoted to the achievement of mobility on a soft lander. But to our knowledge, little attention has been given as to whether the lander should conduct a random walk or whether it should be directed to some target preselected from orbit. We opt for the latter. If the latter, then, one must estimate how much mobility is required and whether the required mobility is feasible. The decision may be in favor of the random walk approach, but, if so, it should be as a consequence of deliberation and not by default.

With respect to penetrators, there is an urgent need to determine whether the considerable potential of these devices can be translated into specific instruments that will adequately characterize samples of Martian subsurface.

Whether the samples are characterized by a soft lander or hard lander, the extent to which they possess reduced carbon, water, soluble electrolytes, gas disequilibria, and ¹³C and ³⁴S depletion should determine the priority accorded to the initiation of a subsequent phase in the biological exploration of Mars, namely, a detailed examination of samples for direct evidence of current or former life.

E. Direct Examination of Samples for Evidence of Current or Former Life

There are two options for conducting the detailed examination: One would be to experiment remotely on the Martian surface; the other is to return samples to Earth and experiment on them here. We believe the arguments strongly favor sample return.

1. Evidence for current life. One second-generation instrument capable of chemically characterizing samples and searching remotely for existing Martian life is currently under development²⁶⁻²⁸—the so-called "Unified Biology Experiment" (now termed the "Integrated Chemistry and Biology Instrument"). The instrument is essentially a chemistry laboratory preloaded with a variety of reagents that can be added to Martian samples in desired sequences or amounts. It uses a mass spectrometer to monitor the gaseous products resulting from metabolism or chemical reaction. As indicated on p. 15, the instrument as presently envisaged would be capable of determining the presence of organic compounds and perhaps characterizing them to some extent, and of measuring gas disequilibria and carbon and sulfur isotope

*In emphasizing these contrasting problems, we are not of course intending to minimize the necessity for intensive study of soft lander payloads.

ratios. It would provide some measure of the amount of water present, but little information on the phases present at subzero temperatures.

Like the existing Viking instruments, it would use metabolic activity as the chief criterion of life detection. But in our view there is an inherent difficulty in demonstrating that data from metabolic experiments which are consonant with biological activity are in fact uniquely ascribable to biological activity. The basic premise of metabolic experiments is that organisms modify substrates and other components of the environment by heat-labile catalysts. While this is true, the modification of substrates by heat-labile catalysts is not unique to living forms. Furthermore, of necessity, most metabolic experiments tend to be highly geocentric. Thus two of the three current Viking experiments are in fact terrestrial microbial experiments in which soil samples are exposed to terrestrially oriented organic substrates under terrestrial conditions of temperature and water. Proposed second-generation experiments are of necessity also geocentric (e.g., challenge soils with various substrates and metabolic poisons).

There are, however, other attributes which are generally considered to be characteristic of living systems, for example: motility in the absence of external vectorial forces; increase in size and number, with descendants similar in chemical composition, form, function, and behavior to the parents; conversion of low-molecular-weight organic compounds into high-molecularweight compounds, or in general a reduction in entropy within a delineated compartment; and objects predominantly composed of specific optical isomers.

The detection and assessment of all these characteristics require sufficient "biomass" for *direct* observation or *direct* chemical analysis. Direct detection requires (a) high sensitivity and versatility in analytical procedures, (b) fractionation and concentration of samples, or (c) prior biological amplification of the sample (i.e., growth) sufficient to permit detection. All three are far more readily achievable on samples returned to Earth than on samples examined remotely on Mars.

2. Evidence for fossil life. The question of whether life ever evolved on Mars is of the same order of importance as the question of whether it now exists. Only a returned sample seems capable of permitting the application of the full armamentarium of paleontological and geochemical techniques: detailed microscopic examination of morphology; the detailed determination of isotope ratios of carbon and sulfur; the absolute dating of the samples; the detailed mineralogical composition; and the localization and relative abundances of carbonates, sulfates, sulfides, phosphates, and nitrates. Remote experimentation also would preclude the powerful approach of simultaneously correlating micromorphology with chemical analysis by such methods as combinations of scanning electron microscopy and electron microprobe analysis.

3. Adaptivity. While Viking is remarkably adaptive in comparison with prior missions, its adaptive capabilities pale in comparison with those of human hands and the human brain on Earth. Adaptiveness is especially important in that a search for current or past life is a search for items about which we will know little in advance.

4. Instrument sophistication and obsolescence. The instrumentation that can be brought to bear in repetitive experiments on samples returned to Earth will always represent a far greater array of far greater sophistication than can ever be launched to Mars. Less obvious is the fact, illustrated by Viking, that any instrument that lands on Mars is of necessity some 10 years out of date by the time it arrives. This obsolescence arises, of course, from the need to freeze instrument design far in advance of the flight.

The chief (and perhaps only) scientific argument against a returned sample (apart from back contamination issues) is the danger of alterations in the sample during the return flight. This potential problem will require extensive study. Perhaps it could be obviated by cryogenic storage. Since Martian samples will have been exposed daily to temperatures below 220°K and will have been exposed annually to temperatures as low as 149°K, it is likely that they could be cooled to and maintained at $\leq 120^{\circ}$ K without significant alteration, temperatures which preclude nearly all thermally driven chemical reactions.

These conclusions about the high scientific potential of returning unsterilized Martian samples to Earth reaffirm existing Space Science Board policy^{1, p. 19} and are consistent with the conclusions of several publications, internal reports, and workshops.³²⁻³⁵ The matter of the physical containment of returned unsterilized samples has also been analyzed by these same and other groups. Containment is considered necessary to protect the Martian samples by providing them with a controlled nonterrestrial environment, and to isolate the Earth from possible Martian organisms until scientific evidence accumulates to show that the risk from interaction is nil or vanishingly small. The reports consider such containment technically feasible.

F. Phases of Biological Exploration versus Missions

Our recommended strategy for the post-Viking exploration of Mars consists of, first, an initial phase of exploration to characterize Martian samples with respect to the possession of several attributes of paramount biological importance; second, a decision as to whether the characteristics of the samples are sufficiently promising to warrant the initiation of a succeeding phase of biological exploration; and, if so, the third step would be this succeeding phase of exploration that would examine Martian samples in detail for evidence of current or past life.

There might or might not be a one-to-one correspondence between the

two phases of exploration and the number of missions flown. One could conceive of a single mission that would combine both phases of exploration, but such a combined mission would preclude the intermediate decision step that we are recommending. A minimum of one mission then would have to be devoted to the first characterization phase. If the decision based on that first phase were positive, our strategy would dictate that the next mission initiate the detailed biological study, and that it be a Mars Sample Return (MSR).* This MSR would have to include aspects of the first phase as well, for clearly it will not be able to acquire samples from precisely the same sites and depths as did the preceding mission or missions.

*The current acronym for this mission is MSSR (Mars Surface Sample Return). We rename it MSR because we recommend subsurface samples.

III RECOMMENDATIONS ON POST-VIKING BIOLOGY STRATEGY FOR MARS

1. Viking has neither confirmed nor ruled out current or past Martian life. Organic compounds have not been detected. Although all three biology experiments have yielded signals that indicate chemical activity, the interpretation of the signals remains ambiguous or inconclusive. Abiogenic explanations seem likely for at least two of the experiments and are probable for the third. We believe that it is preferable to predicate future strategy on the assumption that the signals are not biological in origin.

2. We recommend that the next phase in the biological exploration of Mars should be to acquire and characterize soil samples from areas likely to contain sediments and from ice-regolith interfaces. Locating these areas and locating sites that are shielded from the powerful atmospheric ultraviolet radiation and the powerful surface oxidants will require subsurface sampling.

3. Subsurface sampling can be achieved by soft landers or by hard landers such as penetrators. The choice between the two modes and the attendant costs depend in part on the sampling depths that have to be attained. The required depths are currently unknown. We urge NASA to conduct intensive studies to estimate these depths from data returned from the current Viking and from observations from Earth.

4. The samples acquired from the subsurface of Mars should be characterized with respect to organic compounds, carbon and sulfur isotope ratios, the amount and state of water, the presence of water-soluble electrolytes, and the existence of nonequilibrium gas compositions.

5. Several of the sample characterizations that we consider of paramount biological importance are also considered to be of major geochemical importance by the Space Science Board's Committee on Planetary and Lunar Exploration (COMPLEX).

6. The acquisition and characterization of samples could be carried out by soft landers, by hard landers, or by both. Soft landers can carry the instruments necessary to characterize the samples, but there are serious questions as to whether the landers can reach most of the areas on the surface of Mars that are prime candidates for sampling. Hard landers can probably impact on these prime areas, but there are serious questions as to whether they can carry the instruments necessary to characterize the samples adequately. *We consider*

it urgent that NASA study this paradox in detail to determine how best to resolve it. Its resolution should precede a decision on the next mission.

7. At least one mission would be required for the first, characterization phase of exploration. The greater the extent of reduced carbon, liquid water, soluble electrolytes, and gas and isotopic disequilibrium the subsurface samples possess, the greater the priority that should be accorded the initiation of a second phase of post-Viking biological exploration of Mars—a detailed search for evidence of present or past life.

8. If it is decided to initiate these detailed biological studies, we recommend that they be conducted on samples returned to Earth. We recommend against attempting to perform the detailed studies remotely on the surface of Mars.

9. The current recommendation of the Space Science Board^{1, p. 19} is that "The long-term objectives of exobiology and surface-chemistry investigation are best served by the return of an unsterilized surface sample to earth" and "We, therefore, *recommend* that Mars surface-sample return (MSSR) be adopted as a long-term goal..."

Our recommended strategy is consistent with this policy, but it should be noted that our strategy emphasizes subsurface samples and recommends that the priority accorded biological exploration on a sample return mission should depend on the results of the prior characterization mission or missions.

In 1974 the Board also recommended as preparation for a sample return mission and as an interim cost-effective program "... a Mariner polar orbiter mission and Pioneer survivable hard lander and probe mission...." Although the proposed interim mission would provide invaluable information on geochemical and geophysical characteristics of the Martian surface and interior, either the mission or its scientific payload would have to be supplemented to provide the biological characterizations required prior to decisions on the specifics of the sample return mission.

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