BIOLOGICAL IMPLICATIONS OF THE VIKING MISSION TO MARS

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Abstract. A central purpose of Viking was to search for evidence that life exists on Mars or may have existed in the past. The missions carried three biology experiments the prime purpose of which was to seek for existing microbial life. In addition the results of a number of the other experiments have biological implications: (1) The elemental analyses of the atmosphere and the regolith showed or implied that the elements generally considered essential to terrestrial biology are present. (2) But unexpectedly, no organic compounds were detected in Martian samples by an instrument that easily detected organic materials in the most barren of terrestrial soils. (3) Liquid water is believed to be an absolute requisite for life. Viking obtained direct evidence for the presence of water vapor and water ice, and it obtained strong inferential evidence for the existence of large amounts of subsurface permafrost now and in the Martain past. However it obtained no evidence for the current existence of liquid water possessing the high chemical potential required for at least terrestrial life, a result that is consistent with the known pressure-temperature relations on the planet's surface. On the other hand, the mission did obtain strong indications from both atmospheric analyses and orbital photographs that large quantities of liquid water flowed episodically on the Martian surface 0.5 to 2.5 G years ago.

The three biology experiments produced clear evidence of chemical reactivity in soil samples, but it is becoming increasingly clear that the chemical reactions were nonbiological in origin. The unexpected release of oxygen by soil moistened with water vapor in the Gas Exchange experiment together with the negative findings of the organic analysis experiment lead to the conclusion that the surface contains powerful oxidants. This conclusion is consistent with models of the atmosphere. The oxidants appear also to have been responsible for the decarboxylation of the organic nutrients that were introduced in the Label Release experiment. The major results of the GEX and LR experiments have been simulated at least qualitatively on Earth. The third, Pyrolytic Release, experiment obtained evidence for organic synthesis by soil samples. Although the mechanism of the synthesis is obscure, the thermal stability of the reaction makes a biological explanation most unlikely. Furthermore, the response of soil samples in all three experiments to the addition of water is not consistent with a biological interpretation.

The conditions now known to exist at and below the Martian surface are such that no known terrestrial organism could grow and function. Although the evidence does not absolutely rule out the existence of favourable oases, it renders their existence extremely unlikely. The limiting conditions for the functioning of terrestrial organisms are not the limits for conceivable life elsewhere, and accordingly

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one cannot exclude the possibility that indigenous life forms may currently exist somewhere on Mars or may have existed sometime in the past. Nevertheless, the available information about the present Martian environment puts severe constraints and presents formidable challenges to any putative Martian organisms. The Martian environment in the past, on the other hand, appears to have been considerably less hostile biologically, and it might possibly have permitted the origin and transient establishment of a biota.

1. Introduction

The recent 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 extra-terrestrial 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.

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 discoveries 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 enriched in CO_2) and a surface that is cold, devoid of liquid water, and exposed to highly reactive molecules in the atmosphere and to intense ultraviolet radiation. Earth has an atmosphere intermediate in density, low in CO_2, 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 so as to optimize conditions for its existence (Margulis and Lovelock, 1974, 1977). 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 important questions 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 (Baes *et al.*, 1977). Clearly the stability of equilibria and steady state processes at the Earth's surface and in its atmosphere in the face of 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, studies of biology and of chemical evolution on our neighboring planet could shed important light on these terrestrial questions – questions that could be significant to our ultimate survival.

There have now been numerous reports of the results of the biology experiments proper, but there have been only a few brief analyses published on the biological implications of the findings of the Viking experiments taken as a whole (e.g., Horowitz, 1977). The purpose of the present report, then, is to discuss the biological implications in some detail.

2. Elemental

The discovery of nitrogen in the atmosphere of Mars has eliminated a major barrier to postulating the evidence of a Martian biota. There is general agreement that its absence 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 (Toulmin *et al.*, 1976; Clark *et al.*, 1976). All six elements (and especially phosphorus) are likely to be essential to living systems. Inherent instrument limitations precluded the detection of sodium, but there is no reason to believe it is not present, although probably only in low concentrations (Toulmin *et al.*, 1977).

One striking finding is that the elemental composition of the regolith samples was nearly identical at the two widely separated landing sites (Clark *et al.*, 1976; Baird *et al.*, 1976). This similarity indicates that the fine-grained material in at least the upper surfaces of the regolith has been thoroughly mixed over large regions of the planet – presumably as the result of wind action. It further suggests that the samples analyzed are reasonably representative samples of the fine-grained surface materials over a large part of the planet (Toulmin *et al.*, 1977).

3. Water

A. Water vapour

The atmosphere of Mars has long been known to contain water vapor. Orbital measurements in Viking, however, have shown that there are major geographic,

* Because the word 'soil' implies the presence of organic compounds and connotes a material which 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.

topographic, seasonal, and diurnal variations in the concentrations of atmospheric water vapor, reaching values of 100 precipitable micrometers near the north pole in mid-summer. The relative humidity at the surface is unknown except in the north polar region where orbital observations and calculations indicate saturation (Farmer *et al.*, 1976b, 1977).

B. Ice

The residual north polar cap has been shown to be water ice, probably 1 to 1000 m or more thick (Farmer *et al.*, 1976b; Kieffer *et al.*, 1976), and several experiments confirm or strengthen the inference that large amounts of water are locked beneath the surface in the form of a permafrost that has been present over a considerable portion of the planet's history. First, a number of topographic features photographed by the Orbiters are explicable in terms of the movement of subsurface ice or of its melting and refreezing (Carr and Schaber, 1977). Second, the diurnal behavior of water vapor in the atmosphere indicates that most of it is located near the surface and that at least 80% 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 (Farmer *et al.*, 1976a). The residual north polar cap appears to represent a region where the permafrost 'breaks through' to the surface (Farmer, 1976, personal communication). The third argument for the existence of permafrost follows from analyses of the composition of the atmosphere which suggest 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. One follows from the observed enrichment relative to terrestrial and solar abundances of ^{15}N to ^{14}N (Nier *et al.*, 1976; McElroy *et al.*, 1976; McElroy *et al.*, 1977). If this enrichment has resulted chiefly from preferential escape of the lighter isotope from the upper atmosphere, it indicates that Mars once had at least six times the present abundance of N_2. This same escape process should also have led to an enrichment of ^{18}O which is not observed. To prevent the enrichment from occurring, one needs a large exchangeable reservoir of oxygen, and the most likely reservoir would be water (ice) in amounts equivalent to at least 2 bars of vapor.

A second proposed line of argument (Owen and Biemann, 1976; Biemann *et al.*, 1976a; Owen *et al.*, 1977; Anders and Owen, 1977) follows from the detection of ^{36}Ar, Kr, and Xe in the Martian atmosphere. The amounts of these gases indicate that Mars acquired fewer volatiles during its formation and that the amount of outgassing has been 4 times less than on Earth. Comparison of the ratios of $^{36}Ar/N_2/CO_2/H_2O$ on Earth and Mars indicates that the present Martian atmosphere is deficient in N_2, CO_2, and H_2O. Obtaining a match would require that the planet once had at least fifteen times more CO_2 and five times more N_2 than is now seen in the atmosphere.

Both lines of evidence suggest Mars once had water equivalent to a layer of 10 – 30 m over the entire planet (McElroy *et al.*, 1977; Anders and Owen, 1977).

Amounts equivalent up to perhaps 2 m of global coverage are locked up in residual polar caps; amounts equivalent to some 2.5 m have probably photolyzed and escaped from the planet (McElroy *et al.*, 1977). The rest must be buried in the regolith.

C. Liquid water

Past liquid water. Evidence from orbital photographs is powerful that massive quantities of liquid water once existed and flowed on the Martian surface. Masursky *et al.* (1977) describe three types of water-cut channels, examples of which are shown in Figures 1, 2, and 3. They suggest that the first type, broad channels, (Figure 1) may have been formed when volcanic heat melted overlying permafrost. The second type, sinuous channels (Figure 2), could have arisen by a similar mechanism, but the authors consider a more liely cause to be episodes of climatic

Fig. 1. Broad channel (30 km wide) in the Tiu Vallis area. (Reprinted from Masursky *et al.* (1977), by permission of the American Geophysical Union.)

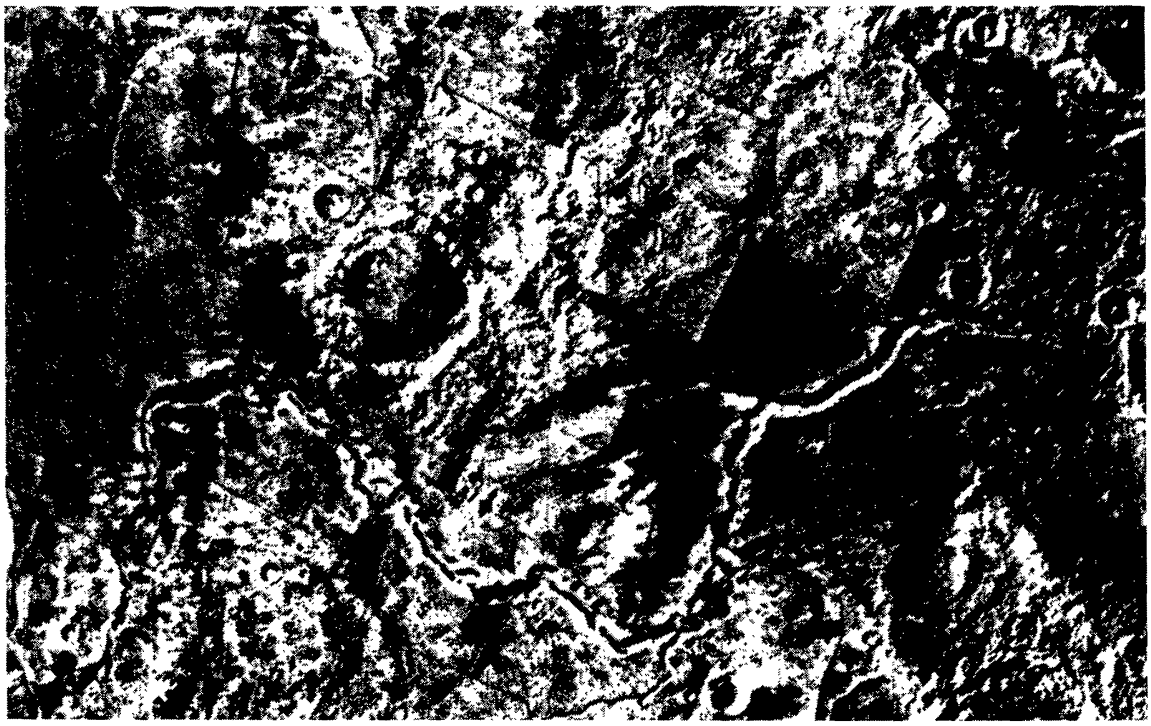

Fig. 2. Sinuous channel in Bahram Vallis due west of the Viking I landing site. (Reprinted from Masursky *et al.* (1977), by permission of the American Geophysical Union.)

warming. The third type, dendritic channels (Figure 3), appear to have been formed as the result of rainfall.

The topographical evidence for surface liquid water in the past is consistent with the conclusions derived from analyses of isotropic ratios of atmospheric nitrogen and argon. These analyses also lead to the conclusion that the total surface pressures of Mars (presently ~7.6 mb (Hess *et al.*, 1977)) exceeded 100 mb in times past (Owen *et al.*, 1977; Anders and Owen, 1977), values high enough to have prolonged the existence of surface liquid water.

Flowing liquid water means the existence of sediments. However, estimates from crater counts suggest that the channels were formed intermittently between 0.5 and 3.5 billion years ago (Malin, 1976; Masursky *et al.*, 1977); hence 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 could be profound. On the other hand, the existence of surface liquid water was probably episodic and its existence may have been too brief to have permitted the origin and establishment of even past life. The volumes of water released during these episodes appear to have been massive ($\geqslant 10^{13}$ m^3), but the water may have evaporated at rates of several to many millimeters per day, and thus could have vanished in a matter of weeks (Masursky *et al.*, 1977). If the released water had stood in open bodies for $\geqslant$ 100 years, it ought to have produced characteristic signs of erosion. But none has been observed. The observational data, however, do not

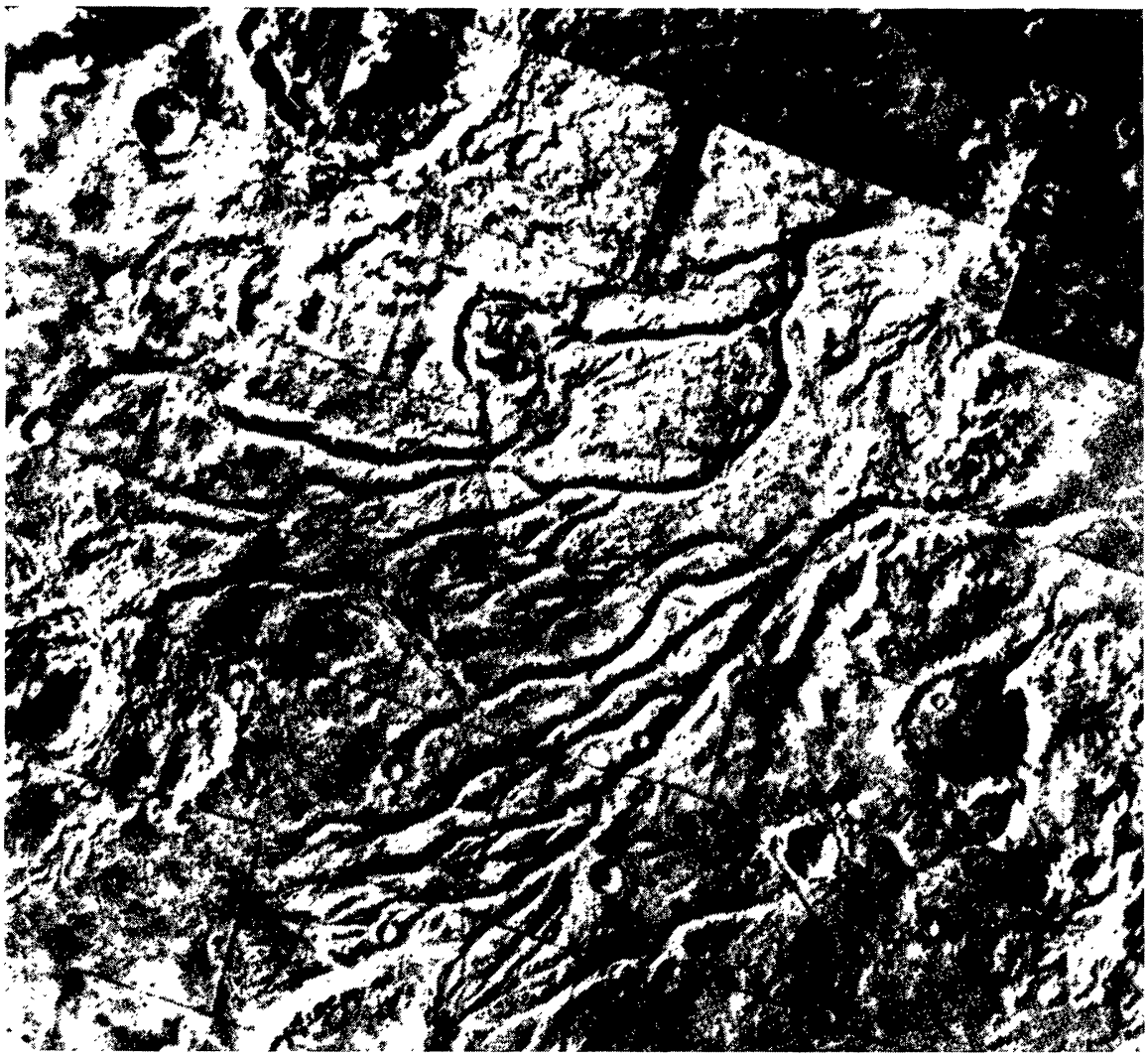

Fig. 3. Two dendritic channel systems draining Lunae Planum west of the VL-1 landing site at Chryse Planitia. (Reprinted from Masursky *et al.* (1977), by permission of the American Geophysical Union.)

exclude the possibility that in local oases a steady production of liquid water counterbalanced the high evaporation rates and permitted the long-term existence of liquid water in a steady state (H. Masursky, 1978, personal communication).

Current liquid water. Liquid water is generally agreed to be essential for the functioning of living forms (see Section 6). But, unfortunately, none of the instruments carried on Viking was designed to detect free liquid water directly. However, liquid water adsorbed to the soil ought to have been detectable in the 200 °C pyrolysis in the Gas Chromatograph Mass Spectrometer (GCMS) of the organic analysis experiment. The GCMS did detect 0.1 to 1 wt % water in regolith samples heated to 350 or 500 °C, but with one exception much less than 0.1% water in samples heated to 200 °C (Table I). The results are consistent with the water being in mineral hydrates of moderate thermal stability (perhaps hydrates of $MgSO_4$) (Toulmin *et al.*, 1976; Clark *et al.*, 1976). The one exception is the sample collected

TABLE I

GCMS estimates of the amounts of water in regolith samples at the two Viking landing sites[a]

Lander and sample	Oven temperature	Amount of water[b]
	(°C)	(wt %)
VL-1		
Sandy Flats	200	<0.1[c]
	500	0.1–1.0[c]
Rocky Flats	350	0.1–1.0[c]
	500	0.1–1.0[c]
	500	0.1–1.0[c]
VL-2		
Bonneville	200	0.05
	350	0.3
	500	1.0
	500	0.25[c]
Under Badger Rock	50	<0.01
	200	0.2
	350	0.3
	500	0.8
	500	0.6[c]

[a] Adapted from Table IV of Biemann *et al.* (1977).
[b] The estimates are based both on the Viking data *per se* and on subsequent laboratory simulations on an instrument with characteristics matching those of the flight instrument.
[c] $^{13}CO_2$ was the carrier gas in these experiments; hydrogen was the carrier in the other experiments. For the reasons discussed by Biemann *et al.* (1977), this difference made the water results less precise for VL-1 than for VL-2.

by the second lander (VL-2) from beneath Badger rock. It yielded no water when heated to 50 °C but about 0.2 wt % when heated to 200 °C. This could represent tightly adsorbed water or mineral hydrate water of low thermal stability.

Toulmin *et al.* (1977) suggest that the apparent cementing of fines into pebble-like aggregates is consistent with the presence of films of adsorbed water around rock particles, films which leached soluble ions from the particles to form the cements – probably sulfate and chloride minerals.

In terrestrial clays, a concentration of adsorbed water of 0.2 wt % would be about 1/50th of that required to form a stable monolayer (Forslind and Jacobsson, 1975). Its water activity (a_w) near 20 °C would be very low ($\ll 0.1$) (Anderson, 1967). (Water activity is a measure of the chemical potential of water. It is equal to p/p_0 where p is the partial pressure of water for the system in question and p_0 is the partial pressure of pure water at the same temperature.)

The detailed measurements of surface temperatures and atmospheric pressures continue to preclude the existence under equilibrium conditions of liquid water with an a_w near 1. The three possibilities for liquid water proposed prior to Viking still remain remote possibilities. (1) Liquid water which has its activity reduced by adsorption to soil. (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 heat (Farmer, 1976). (3) Water that has its activity (and hence freezing point) lowered by the presence of dissolved solutes. This third possibility has been suggested by Anders and Owen (1977) and has been enhanced by the X-ray fluorescence detection of elements like Ca, Mg, Cl, and S which are capable of giving rise to water soluble ions (Toulmin *et al.*, 1976; Clark *et al.*, 1976). (In fact, the existence of $MgSO_4$ at the two landing sites is now considered likely.) Salts like $CaCl_2$, $MgCl_2$, and K_2CO_3 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 pure liquid water in an equilibrium, steady state, or metastable state in the regolith, the surface and subsurface temperatures that have been estimated from orbital infra-red measurements, in conjunction with the low atmospheric pressures, continue to make the likelihood remote. As shown in Table II, summer surface temperatures at the landing sites vary diurnally between $-88\,°C$

TABLE II

Estimates from the infra-red thermal mapper (IRTM) of the maximum and minimum temperatures at the surface at the landing sites and 24 cm below the surface

Site	Season	Diurnal	Depth (cm)	
			0	24
VL-1	Summer–Autumn	Maximum	−3	
				−55
		Minimum	−88	
	Winter	Maximum	−22	
				−69
		Minimum	−95	
VL-2	Summer–Autumn	Maximum	−8	
				−51
		Minimum	−87	
	Winter	Maximum	−82	
				−108
		Minimum	−124	

Data are from Kieffer (1976). The surface temperatures estimated by the IRTM from orbit (resolution ≤8 km) are consistent with the temperatures measured directly by thermocouple in the meteorology experiment 1.6 m above the surface (Hess *et al.*, 1977).

and −3 °C. Some 24 cm below the surface the summer temperatures are expected to be much less variable; i.e., −51 to −55 °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 and −82 °C. Some 24 cm below the surface the winter temperatures at VL-1 and VL-2 will be −69 and −108 °C, respectively (Kieffer, 1976). Mechanisms for providing liquid water below −50 °C become increasingly limited; no mechanisms are known to provide liquid water below about −70 °C. Interlamellar layers of water adsorbed on terrestrial soils remain unfrozen to at least −30 °C (Figure 4, and Anderson and Banin, 1975), but the very forces that keep the water unfrozen make it a difficult source for organisms.

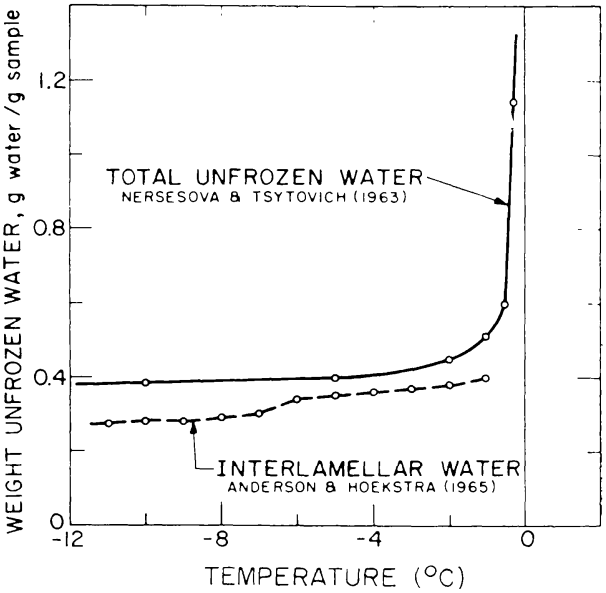

Fig. 4. Amount of total unfrozen water and unfrozen interlamellar water in sodium bentonite as a function of temperature. (Reprinted from Anderson (1967), by permission of Academic Press.)

4. Reduced Carbon and Organics

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 direct evidence of any form of carbon save in the atmosphere and in the winter polar caps. There is, however, indirect evidence that the regolith contains carbonates (Toulmin *et al.*, 1977).

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

* The GCMS estimate for the upper limit for methane in the atmosphere is 5 ppm (T. Owen, personal communication).

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 Bieman *et al.* (1976b, 1977) to be very unlikely. One argument presented is that known terrestrial organic contaminants like 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. Figure 5 shows the gas chromatogram obtained from the pyrolysis of one such antarctic soil sample at 500 °C. The sample is estimated to have contained only 100 living bacteria and a few algae per gram ('rich' terrestrial soils by contrast contain $\geqslant 10^8$ living cells per gram). The highest peak (N – 4) represents benzonitrile at a concentration of 150 ppb. In contrast, no peaks whatever were discernable in the Martian samples, aside from those corresponding to terrestrial organic contaminants.

The concentrations of organics, if any, appear less than the concentration of organics in lunar samples (Murphy *et al.*, 1970; Preti *et al.*, 1971). 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 less (Biemann *et al.*, 1976b,

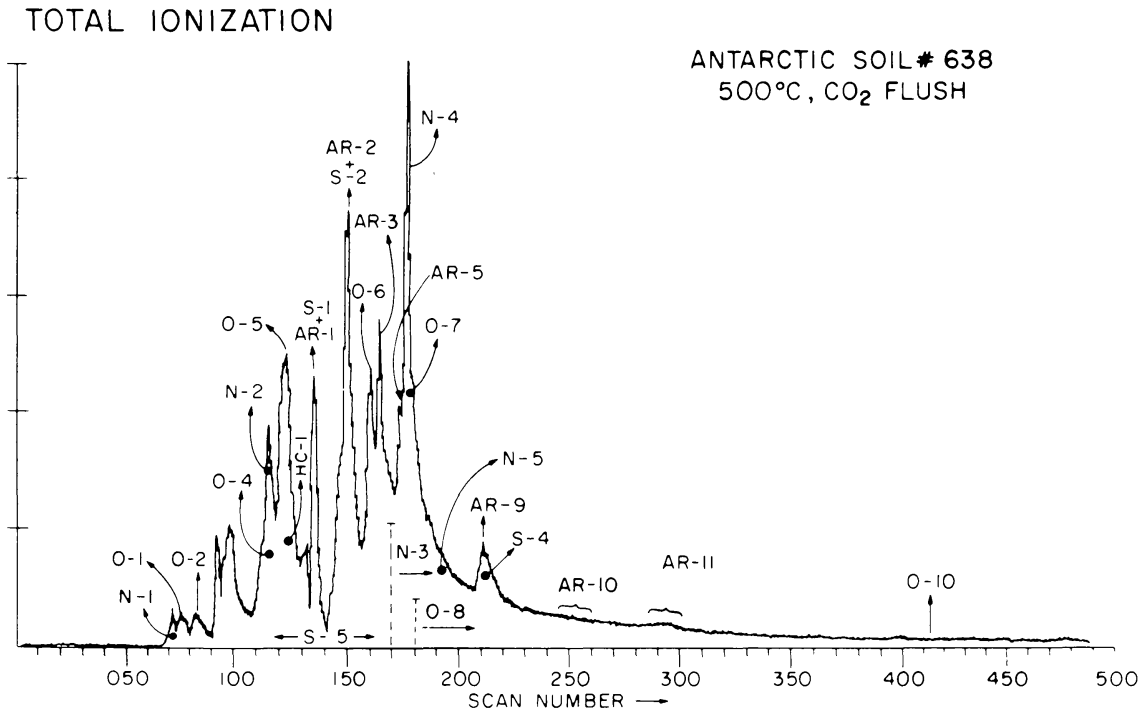

Fig. 5. Gas chromatogram obtained from a sample of Antarctic soil with a laboratory version of the GCMS that corresponds almost exactly to the flight instrument. Each tick mark on the abscissa corresponds to 102.4 s. of elapsed time of the gas chromatogram. The ordinate is linear, with the highest peak (N-4) set at full scale. The compounds corresponding to the identifying codes are given in the original reference. (Reprinted from Biemann *et al.* (1977), by permission of the American Geophysical Union.)

1977)). 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 6 cm 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. They predict the existence of active strongly oxidizing species, especially hydrogen peroxide, near the surface (Hunten, 1974; McElroy *et al.*, 1977). Second, the gas exchange experiment (GEX) on the Viking landers showed the release of up to nearly a micromole of oxygen when the ~1-cc soil samples were humidified with water and warmed to ~10 °C (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 (Bieman *et al.*, 1976b). However, postulating the existence of a viable microbial population under such conditions requires highly 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%, 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% to 0.001% (Baes *et al.*, 1976; Golubic, 1977). Cameron *et al.* (1970) have shown that antarctic soils comparable to that described in Figure 5 contain on the average 10 000 times as much organic carbon as that contained in the living bacteria present in the soils.

If Martain 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%.

5. Biology Experiments

The Biology Experiment consisted of three separate experiments: Gas Exchange (GEX), Labelled Release (LR), and Pyrolytic Release (PR). The first two provided

the Martian soil samples with water vapor at high activity or with liquid water containing organic substrates commonly used by terrestrial microorganisms. The third experiment (PR) provided only two gases known to be constituents of the Martian atmosphere (CO and CO_2), light (optional), and small amounts of water vapor (optional). In all three experiments, the samples were incubated at 8 to 26 °C. The significant measurements were the quantity of gas(es) evolved (GEX, LR, and PR), the type of gas (GEX), or the kinetics of its evolution (GEX and LR). In PR the samples were 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 was 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 indicated that reactants in the Martian samples or the added reactants underwent chemical reactions. The assumption was that microbial activity could be diagnosed from the amounts or types of evolved gases, and from the kinetics of their appearance.

All three experiments yielded signals that clearly indicated chemical activity (Klein *et al.*, 1976), but there is growing evidence that the chemical activity does not represent biological activity. It is true that some aspects of the data are consistent with those expected from biological activity comparable to that observed on Earth. One such instance is the production in the LR experiment of ^{14}C gases when regolith samples were initially moistened with a nutrient medium (Levin and Straat, 1976). A second instance is the synthesis in the PR experiment of picomole quantities of organic matter during the 120-hr incubation of samples in the light (Klein *et al.*, 1976; Horowitz *et al.*, 1977). (However, although statistically significant, the amount synthesized in the PR experiment is only about one-tenth that synthesized by terrestrial soils that gives the minimal observed response, i.e., antarctic soils (Horowitz *et al.*, 1972, 1976).) Thirdly, in the LR experiment the activity was abolished when the regolith samples were preheated to 160 °C for 3 hr. Heating to such temperatures, of course, also abolishes biological activity in terrestrial samples.

But there are three major difficulties with a biological interpretation of the data: The evolution of oxygen in the GEX experiments, the response to heating in the GEX and PR experiments, and the response to water in all three experiments.

(1) *The evolution of oxygen in GEX.* In the GEX experiment, the collected samples produced no detectable gas changes until they were humidified, but humidification produced a relatively rapid evolution of oxygen (Figure 6). The evolution of oxygen occurred in all three samples from both landing sites, including the sample at VL-2 obtained from underneath Notch rock (Table III). It reached a maximum value in one run of nearly a micromole. Oyama and Berdahl (1977) report that, in pre-Viking simulation of the GEX on terrestrial soils, they never observed oxygen evolution in the dark.

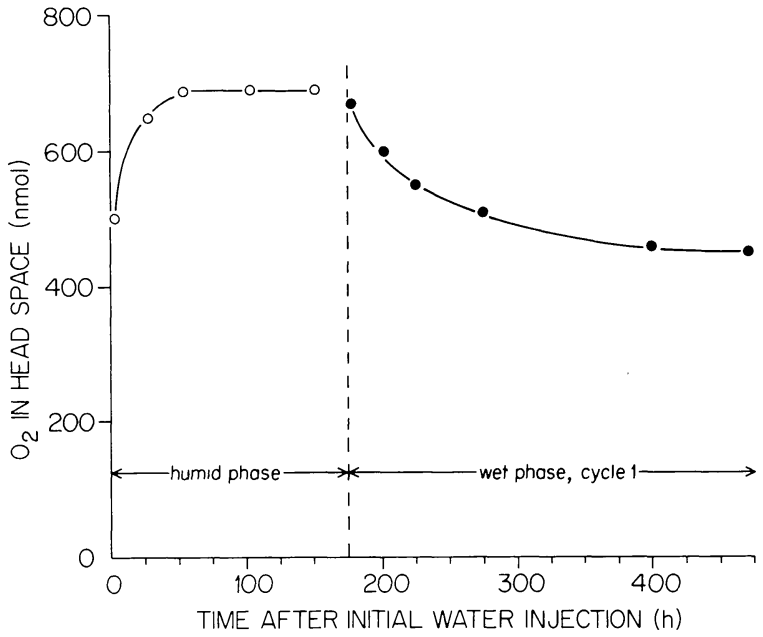

Fig. 6. Amount of oxygen in the head space of the GEX experiment as a function of time after the initial introduction of 0.57 ml of nutrient medium to a 1 cm^3 soil sample from VL-1 (Chryse, Sandy Flats). Some 175 h later an additional 2.3 ml of medium was added to terminate the 'humid phase' and initiate the 'wet phase' (see text). The incubation temperature during the humid phase was 8.3 to 11.6 °C (mean 10.3 °C). The amount of oxygen has been corrected for dissolution of gas in the added liquid and several other factors (see original reference Table III). The authors calculate that to the plateau value of 690 nmoles must be added 85 nmoles associated with the oxidation of ascorbate in the medium. (Drawn from data of Oyama *et al.* (1977).)

TABLE III

Total amount of oxygen evolved in the gas exchange (GEX) experiment after humidification of regolith samples[a]

Lander and sample	Oxygen in head space		
	Prior to humidification	After humidification	
		Observed	Corrected[b]
	(nmol)	(nmol)	(nmol)
VL-1			
Sandy Flats	4	690	775–790
VL-2			
Beta	4	110	190
Under notch rock[c]	4	70	70–270

[a] From data of Oyama *et al.* (1977) and Oyama and Berdahl (1977).
[b] The chief correction was for the oxygen absorbed by the ascorbate in the medium. The 'observed' values in the prior column were adjusted for losses of gas during sampling, for pressure changes, and for dissolution of gases in the aqueous medium.
[c] This sample was wet with nutrient medium without being first humidified.

(2) *The response to added water.* A second major difficulty with biological interpretations of the data is the response of samples to water. In the GEX experiments, the samples were first humidified by adding ~0.5 ml (0.49–0.59 ml) of the complex aqueous nutrient medium to the bottom of the 8.7 cm^3 test cell in such a way that the 1 cm^3 sample was contacted only by water vapor and not by the liquid medium. After following the gas changes for seven Martian days (sols), an additional 1.8–2.3 ml of medium was added, an amount sufficient to wet the sample with liquid. Some 13–78 sols later a third ~2-ml aliquot was added. In the LR experiment, 0.115 ml of liquid medium was added initially to 0.5 cm^3 of sample, and the liquid contacted only the central core of the sample. Subsequent additions of medium wet the entire sample. The PR experiment was run either without the addition of any water or with the addition of about 80 μg of water *vapor* to 0.25 cm^3 of soil in the 4 cm^3 test cell.

In the earlier testing of the Viking biology experiments on terrestrial soils, the presence or addition of liquid water was required for the manifestation of microbial activity (Horowitz *et al.*, 1972; Oyama *et al.*, 1972; *Levin et al.*, 1972), and the expectation in at least the LR and GEX experiments was that, if signals represented Martian biological activity, that activity would be enhanced or sustained first by the introduction of liquid water and secondly by the addition of the nutrients contained in the water. But the Viking results are not consistent with these expectations. In GEX, as shown in Figure 6, all of the oxygen that was evolved during the initial humidification cycle when the samples were in contact only with water vapor. The actual wetting of the samples with liquid medium in fact produced a slight *decrease* in the amount of oxygen in the head space of the test cell, presumably as the result of quantities being absorbed by the additional ascorbate added to the system. Analogous results were observed in the LR experiment (Figure 7). Nearly all of the evolution of ^{14}C gases (presumably chiefly $^{14}CO_2$) occurred after the first addition of liquid medium (which, as noted above, was designed to come in contact only with the central portion of the sample). Subsequent additions of liquid medium produced an abrupt *decrease* in ^{14}C gases in the head space followed by a slow further evolution at a seemingly constantly decreasing rate. Levin and Straat (1977) suggest that the

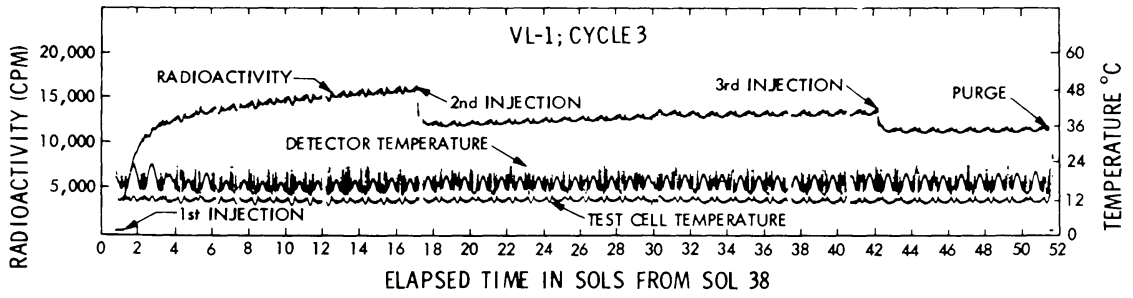

Fig. 7. Kinetics of evolution of ^{14}C gases in the LR experiment when a sample from VL-1 (Sandy Flats) recieved three sequential injections of nutrient medium (see text). (Reprinted from Levin and Straat (1976), by permission of AAAS.) The radioactivity released corresponds to about 30 nanomoles of CO_2 cc^{-1} of sample (Klein, 1977).

abrupt decrease may reflect dissolution of gas in the added aqueous nutrient, perhaps aided by changes in pH produced by the soil wetting.

In other words, in the LR experiment the initial addition of medium, which wet only a portion of the sample, appeared nearly to exhaust the reactants in the entire sample. In the GEX experiment, the initial introduction of water *vapor* completely exhausted the reactants that were the source of the oxygen, i.e., further additions of liquid aqueous medium produced no further evolution of oxygen.

The effect of the introduction of water vapor into the samples in the PR experiment is far different from that observed on terrestrial samples, and the effect is difficult to reconcile with a non-terrestrial biology. In prior simulation with terrestrial soil samples, a positive PR response required the addition of water. The effects of water on the Viking samples are summarized in Table IV. Here we see that the addition of water either had no effect on the size of Peak 2 (which represents the amount of organic material synthesized during the course of the experiment) (Sample C-6) or it abolished Peak 2 (Sample U-2). Note that the sample from under Notch rock also yielded a second peak that was not significantly different from zero. This may have been due in part to the absence of light and in part to a higher water content in the sample. (Recall that the GCMS found that the

TABLE IV

Summary of results of Viking pyrolytic release (PR) experiment[a]

Lander and sample	Heat treatment	Light	Added water	Disintegrations per minute	
				1st peak[b]	2nd peak[c]
VL-1					
C1 Sandy flats-1	No	Yes	No	64 464	842 ± 29
C3 Sandy flats-2	No	Yes	No	61 027	214 ± 28
C4 Sandy flats-3	No	Yes	No	18 545	289 ± 31
C6 Sandy flats-3 (stored 139 sols)	No	Yes	Yes	193 803	255 ± 31
C2 Sandy flats-1 (stored 19 sols)	175 °C	Yes	No	69 536	105 ± 29
C5 Sandy flats-3 (stored 69 sols)	90 °C[d]	Yes	Yes	20 295	275 ± 29
VL-2					
U-1 Beta 1	No	No	No	64 845	178 ± 31
U-2 Beta 2	No	Yes	Yes	113 845	−7 ± 28
U-3 Under notch rock	No	No	No	118 309	36 ± 35

[a] Modified from Table I of Horowitz *et al.* (1977).
[b] Material not adsorbed by organic vapor trap (OVT).
[c] Counts of material retained on OVT corrected for contamination from peak 1. This peak represents the CO or CO_2 fixed as organic material during the course of the incubation.
[d] See text.

sample from under Badger rock contained appreciably more 'loosely' attached water, i.e., water driven off at 200 °C, than the samples from open areas (Table I).)

It needs to be emphasized that the physical-chemical status of added water in the PR experiment is very different from that in the GEX and LR experiments. Toulmin *et al.* (1977), on the basis of computer matching of the elemental abundances in Martian samples against numerous terrestrial minerals, believe it likely that Fe-rich montmorillonite clays are a major component of the Martian fine-grained material. Figure 8, adapted from Anderson's (1967) review, shows water

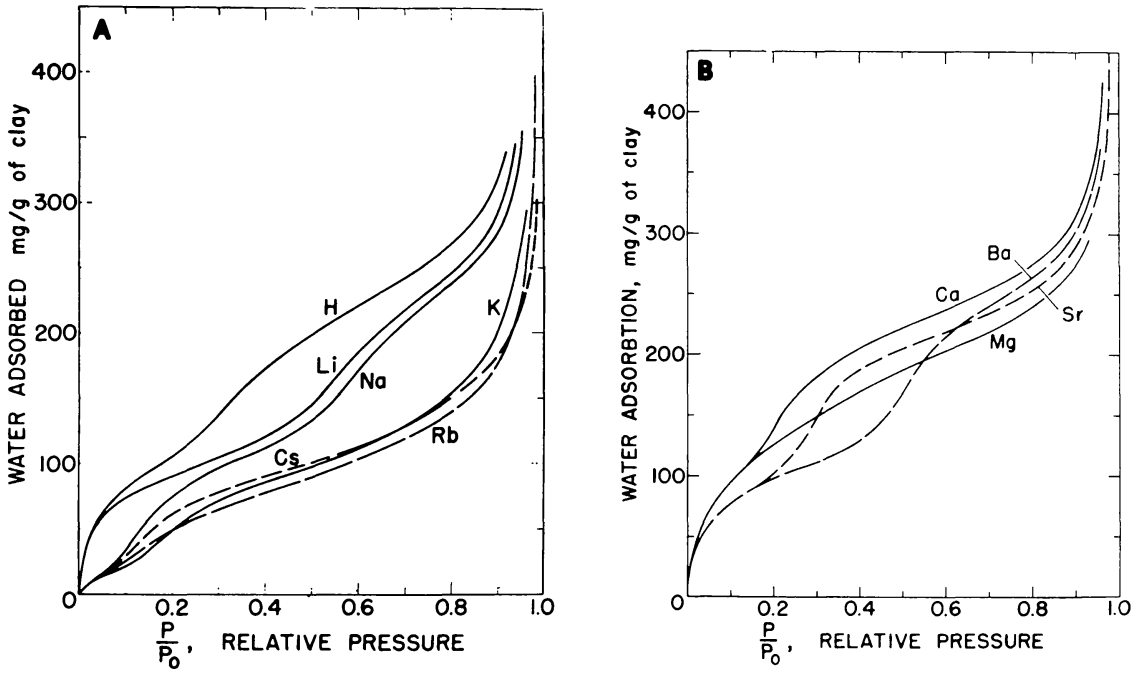

Fig. 8. Water desorption isotherms of homoionic montmorillonite saturated with various monovalent cations (A), or divalent cations (B). (From Anderson (1967) by permission of Academic Press.)

adsorption isotherms for montmorillonite saturated with various monovalent and divalent cations. If we assume that the adsorption isotherms of Martian materials are similar* and if we assume a bulk density of 1.5 for the sampled material,** then the ~ 0.5 ml of water initially added to the 1 cm^3 sample in the GEX experiment would have produced a water activity ($a_w \equiv p/p_0$) of > 0.9, provided that the 7 sol incubation permitted the water vapor to diffuse to equilibrium. The subsequent additions of aqueous medium would have completely saturated the system and produced an a_w close to 1.0. In the LR experiments, the initial addition of 0.12 ml medium to the central portion of a 0.5 cm^3 sample ought to have produced an

* The assumption is not critical. The adsorption isotherms for materials as disparate as montmorillonite, DNA, protein, lecithin, and whole bacterial cells are remarkably similar (Falk *et al.*, 1962; Bull *et al.*, 1964; Elsworthy, 1961; Bateman *et al.*, 1962). They fall within the bounds of the curves in Figure 8.

** Shorthill *et al.* (1976), give values of 1.0 to 1.8 g cm^{-3}.

immediate water activity close to 1.0 in that central portion, but ultimate equilibration throughout the sample would have resulted in a water concentration of ~ 0.16 g H_2O g^{-1} of sample and an a_w of ~ 0.2 to 0.8. In the PR experiment, the ~ 80 μg of water added to 0.25 cm^3 of soil would upon equilibration have produced an equilibrium water concentration of ~ 0.0002 g H_2O g^{-1} sample, which for montmorillonite would result in a water activity very close to zero. In fact, this amount of added water (0.02%) is probably less than that present in the indigenous Martian material (cf. Table I). The a_w's corresponding to the water contents in the PR experiment are far below the lowest value of 0.61 ever recorded for a terrestrial organism (see Section 6).

These rough estimates of the equilibrium water activities in samples during the experiments are summarized in Table V. The values are not likely to be significantly affected by the involvement of water in the chemical reactions observed in the samples, for the molar ratio of added water to that of the observed products far exceeded 1000. Not only do the equilibrium values differ appreciably, but the kinetics of the approach to equilibrium must have differed greatly both among the three experiments and also in different regions of a sample in a given experiment.

TABLE V

Estimated equilibrium water activities in regolith samples in the Viking biology experiments

	Equil. a_w after	
Experiment	Initial addition of water	Second addition
GEX	>0.9	~ 1.0
LR	0.2 to 0.8	~ 1.0
PR	$\sim \ll 0.1$	–

(3) *The responses of heated samples.* To test any putative biological activity, the biology experiments provided the option of heating the samples of regolith to as high as 180 °C before conducting the experiment at ≤26 °C. In the LR experiment, heating to 160 °C eliminated all subsequent activity and heating to 46–50 °C reduced activity by about two-thirds (Figure 9). However, in the GEX and PR experiments the reactants did not appear to be nearly so thermally labile. In the GEX experiment, different results were obtained for the two samples heated to 145 °C. A sample from VL-1 Sandy Flats evolved 250 nmoles of oxygen after subsequent humidification, a value that was about a third of its unheated counterpart (Table III). The other heated sample (from VL-2 Beta), however, evolved no oxygen after heating. Oyama and Berdahl (1977) suggest that the vent tube was

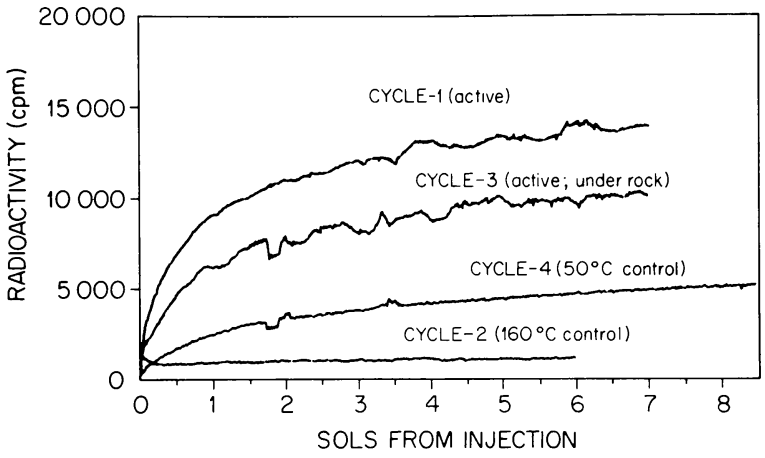

Fig. 9. Kinetics of evolution of ^{14}C gases after various regolith samples in the LR experiment had received aqueous nutrient medium. The samples represented by the upper two curves were collected from the open and from under Notch rock, respectively. The samples represented by the lower two curves were heated to 46° and 160 °C for about 3 hr, respectively, and then allowed to cool prior to the injection of water at ≤18 °C. The bottom curve is from VL-1; the other three curves are from VL-2. (Replotted from Levin and Straat (1977).)

blocked in this latter case, and this would have caused any water vapor driven off during the heating to have remained in the headspace.

In the PR experiment, prior heating of a sample to 175 °C for 3 hr (Sample C-2, Table IV) reduced the size of the second peak to 12 to 50% of that in comparable unheated samples (C-1, -3, -4). But heating to 90 °C for 112 min produced no decrease in subsequent activity (C-5 versus C-4 and C-3). Samples C-4, -5, and -6, the last to be carried out, were all performed on aliquots of a sample collected by the VL-1 lander and stored at 5 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. As mentioned, the response of this last sample differed from the results of the previous sample (U-2) run in the presence of water, a run which had given a second peak of zero.

The rather wide differences in thermal stability observed both among GEX, LR, and PR and within some of the experiments could possibly be the result of differences in the water contents of the samples. Thermal *lability* is explicable in either chemical ot biological terms. However, we find it difficult to believe that thermal *stability* at 175 °C for 3 hr is consistent with the survival of biological systems. It certainly is not on Earth.

A. Possible non-biological explanations for viking biology results

Intensive attempts are now being made to simulate the results of the Viking biology experiments abiologically. Although the information at this writing is preliminary,

major features of the LR and GEX experiments have been mimicked at least qualitatively by non-biological reactions. The major feature of the LR experimental results is almost certainly 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 (Vol'nov, 1966; Ponnamperuma *et al.*, 1977).

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 (Ponnamperuma, 1977; Wydeven *et al.*, 1976, personal communication). Wydeven and his colleagues have also obtained oxygen evolution in amounts and at a rate comparable to that observed in GEX on Mars by exposing soil 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 (Hunten, 1974). Splitting of H_2O gives H and OH which lead by well-characterized pathways to the production of H_2O_2.

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 H_2O added) were irradiated at >320 nm (0.5% $<$ 320) in the presence of $^{14}CO_2$ and ^{14}CO, and then pyrolyzed at 625 °C, significant quantities of ^{14}C–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 $^{14}CO_2$ 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 $^{14}CO_2$ and ^{14}CO (less than 0.01% 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 (Horowitz *et al.*, 1972).

It is known, however, from the work of Hubbard *et al.* (1971, 1973) that non-biological organic synthesis can occur under conditions analagous 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, CO_2, and small amounts of H_2O 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 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 (Hunten, 1974). Hydrogen peroxide dissociates into hydroxyl radicals when irradiated with even visible light (quantum yield 0.3 at 313 nm and measureable reactivity at 365 nm

(Calvert and Pitts, 1966)). And hydroxyl radicals are believed to play a role in the abiogenic syntheses observed by Hubbard *et al.* (Tseng and Chang, 1975).

Possibly then the reaction occurring in the PR experiment on Mars is something akin to $H_2O_2 + CO \xrightarrow{h\nu}$ formic acid. (This illustrative reaction is at least thermodynamically feasible, for it has a free energy of -20 kcal mole^{-1}. Synthesis along this pathway, however, would require special conditions, for the degradative oxidative pathway, which yields CO_2 and H_2O, has a free energy of -90 kcal mole^{-1} (N. H. Horowitz, 1976, personal communication).)

We are not proposing that this is necessarily one of the actual reactions that occurred in the PR experiment. (For example, the role of light is not clear. The two samples incubated in the dark (Table IV) gave low second peak counts, but not counts of zero. For other speculations on mechanisms, see Oyama *et al.* (1977).) 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. Nevertheless, in our view the weight of the evidence is that the positive signals from the biology experiments are not biological in origin. That judgment rests chiefly on seven points. (1) The direct evidence from GEX and indirect evidence from other experiments and modeling that the regolith contains strong oxidants, (2) the inhibitory or dissipative effects on the reactions of the presence of added water, especially when the a_w was $\geqslant 0.2$, (3) the ability of reactants in the PR experiment and in one of the GEX runs to continue to function after being heated to $\geqslant 145$ °C, (4) the lack of detected organic compounds, (5) the ability to account, at least qualitatively, for the results of GEX and LR by nonbiological reactions, (6) 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 (7) the existence of at least conceivable mechanisms for a different wavelength dependence at the Martian surface.

These conclusions apply to the specific data obtained in a small number of samples at two highly localized but widely separated spots on the planet's surface. But what can one infer from the Viking results about the likelihood of present Martian life elsewhere in the planet, and what can one infer about the possibility that life evolved in times past but no longer exists?

In assessing the former possibility, it would be helpful to examine how terrestrial organisms would be expected to fare on Mars. It is not that the limits to terrestrial growth are necessarily the limits to other forms of life, but that a comparison between the known enviromental limits for terrestrial organisms and the known

and estimated enviroments on Mars should help clarify the constraints and requirements that would be applicable to a Martian organism. Furthermore, the question of the likelihood of the growth of terrestrial cells on Mars is of direct concern to what quarantine steps will be required in follow-on missions to Mars.

6. Environmental Limits to the Growth of Terrestrial Microorganisms and Its Relevance to Mars

Table VI summarizes the limit for the growth of terrestrial organisms and compares these limits with those that prevail on Mars. Although orbital measurements have covered appreciable fractions of Mars' surface, the two Landers have sampled only a few square meters of the surface at two widely separated subpolar sites. The biologically relevant experiments were conducted on soil samples acquired during the Martian summer and early fall from as deep as 6 cm below the surface. Nevertheless, certain extrapolations relevant to the question of the growth of

TABLE VI

Conditions on Mars versus approximate limits for the growth of terrestrial microorganisms

Factor	Conditions on Mars[a]	Limits for growth of terrestrial organisms	Comments and references
Water activity (a_w)	0 to 1	≥ 0.83	For bacteria and yeasts. Limit for fungi is ≥ 0.61 (Rose, 1976).
Water liquid	Not detected	Required	
Temperature	+20 ° to −143 °C	> −14 °C	See text and Figure 10.
pH	Not known	< 11.5; > 0	Deal *et al.* (1975); Margulis *et al.* (1977).
UV radiation	0.04 joules cm^{-2} min^{-1}[b]	0.1 joules cm^{-2}[c]	Donnellan and Stafford (1968); Margulis *et al.* (1977).
Ionizing radiation	< 500 rad yr^{-1}[d]	2–4 m rad[c]	Goldblith *et al.* (1953), Kirk and Othmer (1969), Silverman and Sinskey (1968).
Nutrients	Organic compounds ≤ ppb; most required elements detected	specific requirements for both types and concentrations	Margulis *et al.* (1977)
Antimetabolites	Strong oxidants present	None present	

[a] See text except where noted.
[b] Averner and MacElroy (1976).
[c] Limits for survival; limits for growth are not known.
[d] The chief contributors to ionizing radiation would be protons from galactic cosmic rays and solar flares. The flux from the latter far exceeds that from the former, but the great bulk of the solar protons have energies ≤ 1 MeV, and such protons can penetrate < 0.1 mm of material with a density of 1. The value of < 500 rad yr^{-1} assumes that such shielding is present. The calculations were made by M. L. Randolph (Oak Ridge National Laboratory) on the basis of flux data supplied by C. W. Snyder. We appreciate their assistance.

terrestrial organisms can be made with various degrees of confidence to other regions of the planet, to greater depths, and to other seasons of the year.

We will first compare terrestrial growth limits to the conditions that were observed in the upper 4–6 cm at the landing sites, and will then extrapolate successively to the upper layers of the subpolar regions as a whole, to subsurfaces in the subpolar regions, and finally to the polar regions covered by the residual water ice cap.

A. Conditions at the Landing Sites in the Upper 4–6 cm of the Regolith

(1) *Water.* Although water exists on Mars in the form of ice and vapor we have already discussed the reasons for believing that there is little probability of the existence of liquid water at high activity anywhere on the planet. Both aspects of water (the liquid state and high activity) are required for the functioning of living terrestrial organisms. There is no convincing case of cellular function and growth in the presence of water vapor alone, regardless of its activity; and in the presence of liquid water, the minimum recorded water activity for growth is 0.61 for a fungus. The great majority of terrestrial microorganisms, however, require water activities >0.9 (Rose, 1976). Water activities do reach 1.0 on Mars, but only at temperatures far below those that permit the existence of pure liquid water. The GCMS experiments showed that 0.05 to 0.2% of the Martain samples was water that was capable of being released by heating to 200 °C (Table I). But such amounts of water in clays at 0 to 20 °C would have activities far less than 0.1.

Another argument against the existence of high activity liquid water at the landing site is the findings of the LR and GEX biology experiments. In both cases, the initial addition of water vapor or liquid water to the soil samples in quantities sufficient to raise the a_w to 0.2 to 0.9 dissipated the reactants so that further additions produced no further reactions (release of ^{14}C gases and release of oxygen in the two experiments, respectively). Presumably, therefore, no reactions at all would have been observed if the soil itself had been exposed to high activity liquid water just prior to the acquisition of samples by the Landers.

(2) *Temperature.* The maximum temperature observed to date at the surface of the landing sites during the summer-autumn observation period is -2 to -3 °C. This is below the minimum growth temperature of most terrestrial microorganisms, although, as we shall discuss shortly, a few terrestrial organisms can grow at temperatures as low as -14 °C. (In the southern hemisphere of Mars, the maximum summer surface temperatures may reach 20 °C.) But at night, even in summer, the temperature drops to $\leqslant -83$ °C. Dry bacterial and fungal spores could survive many cycles of such freezing, but hydrated and germinated spores or vegetative cells of most terrestrial species could not (Mazur, 1966, 1968; Leef and Mazur, 1978). And any terrestrial microorganism that is to grow or carry out normal functions on Mars must by definition be in the vegetative state for at least brief periods of time.

(3) *Lack of detected organic compounds.* We have noted that no organic compounds other than traces attributable to terrestrial contaminants were detected in regolith samples analyzed by the GCMS. The inability to detect organic compounds does not, of course, prove that none was present; but even if trace amounts are present, the probability is remote that these would provide a nutrient medium that could be utilized by terrestrial microorganisms (see Margulis *et al.*, 1977, for further discussion).

(4) *Elemental.* Most of the elements critical to terrestrial cells have been detected (carbon, nitrogen, oxygen, calcium, sulfur, magnesium, chlorine, and probably phosphorous and potassium. Sodium was undetectable but is likely to be present). As mentioned, the elemental composition at the two landing sites was close to identical.

(5) *Oxidants.* We have reviewed the evidence that strong oxidants are present in at least the top few centimeters of the regolith at the landing sites. Presumably, the oxidants were responsible for the unexpected inability to detect organic compounds, i.e., they are strong enough to decompose carbon-carbon bonds faster than they accumulate from the infall of carbonaceous chondrites or can be synthesized. The GEX experiment showed that the oxidants were present in the samples collected from beneath Notch rock, a rock which presumably had lain undisturbed for many years. Finally, the experiment showed that the oxidants were present at both landing sites.

While there are highly specialized terrestrial organisms capable of functioning in the presence of moderate concentrations of strong oxidants (e.g., 1*M* surfuric acid), we know of none capable of surviving oxidants powerful enough to decompose organic compounds to the extent that they would be undetectable in a GCMS-type instrument.

B. Extrapolation from the Viking findings to the planet's surface as a whole, to regions below the surface, and to other seasons of the year.

Surface temperatures in the Martain winter will drop far lower than those experienced during the Lander experiments. Estimates from the infra-red thermal mapper (IRTM) indicate that the *maximum* surface temperature will fall below −15 °C (the minimal terrestrial growth temperature – see below) for more than half the Martian year at the VL-2 site (48 °N) and further north. At VL-1 (22 °N) the maximum surface temperature will just about reach −15 °C in the winter (Kieffer, 1976).

In considering extrapolations from the findings of VL-1 and VL-2 on surface chemistry, we note that, although the two landing sites (22 °N and 48 °N) are separated by some 1500 km in latitude and 176 deg in longitude, the results of the GEX and LR biology experiments and of the organic and inorganic analyses at the two sites were either similar or essentially identical.* Strong similarities were

* Samples from the two sites in the Pyrolytic Release (PR) experiment, however responded differently to the addition of water vapor. Horowitz *et al.* (1977) suggest that this reflects differences in the properties of the soil at the two sites, but they draw no inferences as to the nature and degree of the differences.

evident as well from the imaging experiment and from the atmospheric analyses. The results of the GEX, LR, and GCMS experiments are consistent with the presence of powerful oxidants in the surface samples. Since these oxidants are almost certainly derived from atmospheric photochemical reactions or from chemical reactions between atmospheric species and the regolith, there is every reason to expect that they will be globally distributed in the Martian surface, except possibly in the residual polar caps.

Certain extrapolations can also be made to depths below the 4–6 cm sampled by the Landers.

(1) *Water.* We have noted in Section 3 that several Viking experiments confirm or strengthen the inference that large amounts of water are locked beneath the surface in the form of ice. It is conceivable that some of this water is liquid; however, because of the low temperatures at subsurfaces (see below), the existence of bulk liquid water in an equilibrium state would require multimolar concentrations of electrolytes (Table VII).

TABLE VII

Solute concentrations and water activities in NaCl solutions at various temperatures

Temperature (°C)	Concentration NaCl[a] (molal)	a_w[b]
−5	1.45	0.95
−10	2.79	0.91
−14	3.73	0.87
−16	4.17	0.85
−18	4.58	0.84
−20	4.99	0.82

[a] In the unfrozen portion of the solution. From International Critical tables (1926).

[b] $a_w = p_{H_2O}$ (solution)/p_{H_2O} (liquid, pure) ≡ p_{ice}/p_{H_2O} (liquid, pure). Calculated from data of Dorsey (1940). See Figure 11. The activities depend only on temperature (and total pressure); they are independent of the nature of the solute.

(2) *Temperature.* The most thorough review known to us of the minimum growth temperatures of terrestrial microorganisms is that of Michener and Elliot (1964). A histogram summarizing their findings on reports of growth below 0 °C is shown in Figure 10. Many of these reports are based on incubations of over a year. Bacteria are considered separately from fungi because the latter are nearly all aerobic and would be incapable of growing at Martian partial pressures of oxygen (at the surface O_2 is 0.13% of the Martian atmosphere (Owen *et al.*, 1977)). The single case of a bacterium growing below −12 °C was a report of growth at −20 °C. Neither it nor the three reports of fungal growth below −12 °C have been

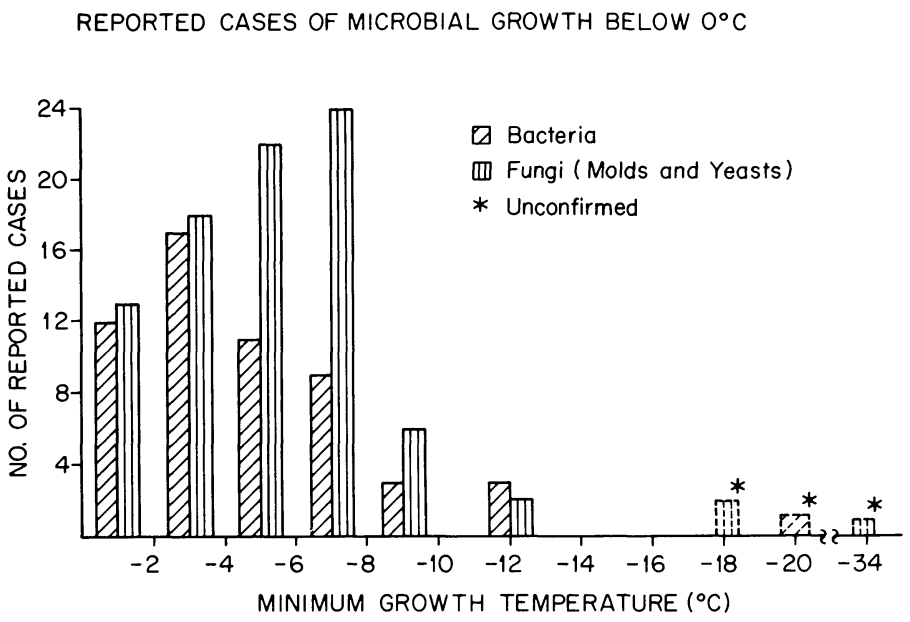

Fig. 10. Reported cases of microbial growth below 0 °C. (Adapted from Michener and Elliott (1964).)

confirmed. Michener and Elliot point out that "The best evidence that growth does not generally occur in foods in this temperatue range (i.e., < -17 °C) is that billions of cartons of frozen food have been stored at or near -18 °C without reported microbial spoilage".

A more recent survey by Fennema *et al.* (1973) confirms Michener and Elliot's conclusion that microbial growth in foods does not occur at -18 °C.

This inability of organisms to grow below -12 °C is consistent with the known physical state of aqueous solutions at these temperatures. As Table VII shows, when solutions of sodium chloride in water, for example, are equilibrated at various subzero temperatures, the concentrations in the unfrozen portions exceed 3.7 molal at and below -14 °C. For solutes in general, the concentrations of solutes in the unfrozen portions of solutions are given by $\phi \nu m = \Delta T/1.86$, where ϕ is the osmotic coefficient, ν the number of species into which the solute dissociates, and m is the molality (Lewis and Randall, 1961). Aside from the toxic effects to nearly all microorganisms of such high concentrations of electrolytes, the high concentrations also depress the water activity (a_w) below the value permitting the growth even at optimal temperatures of all microorganisms save halophilic and osmophilic forms. As shown in Table VII and Figure 11, the values of a_w at -14, -16, -18, and -20 °C are 0.87, 0.85, 0.84, and 0.82, respectively.

At the Viking landing sites, the maximum summer temperatures some 6 cm below the surface are estimated from the orbital infra-red measurements to be -35 °C. This temperature is 20 ° *below* the minimum confirmed growth temperature for terrestrial microorganisms, and it is even below the lowest growth temperature ever claimed in published reports. At a depth of 24 cm, the maximum summer temperatures at the VL-1 and VL-2 sites are estimated to be -50 °C, or

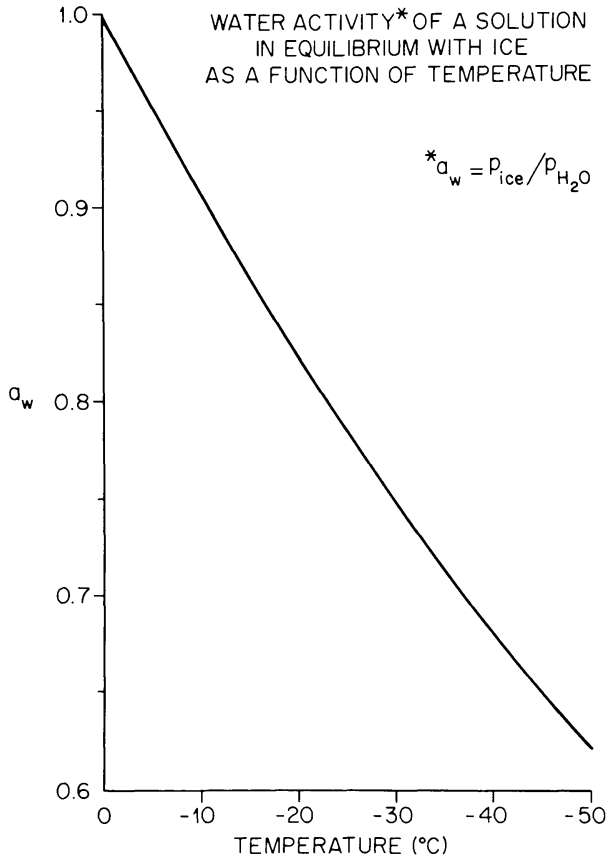

Fig. 11. Water activity (a_w) of an aqueous solution in equilibrium with ice at various temperatures. The water activity equals p_{ice}/p_{H_2O}, where p_{ice} and p_{H_2O} are the vapor pressures of ice and pure supercooled water at a given temperature. The vapor pressure data are from Dorsey (1940), Tables 243, 244, and 264.

35 ° below the minimum confirmed terrestrial growth temperature. In the southern hemisphere as a result of the eccentricity of the Martian orbit, the maximum surface temperatures between latitudes 5 ° and 45 ° are about 15 °C warmer than at the present landing sites (Kieffer *et al.*, 1977). As a result, at subsurface depths sufficient to damp out diurnal variations, the maximum summer temperature is calculated to be about −35 °C, still some 20 °C below the minimum confirmed terrestrial growth temperature.

(3) *Ultraviolet light.* As shown in Table VI, the flux of ultraviolet radiation impringing on the Martain surface would be rapidly lethal to any terrestrial organism. However, the UV flux is sharply attenuated below the surface. For example, Sagan and Pollack (1974) estimate an attenuation of several million-fold at a depth of 0.8 cm.

(4) *Oxidants and organic compounds.* Since the oxidants in the regolith are almost certainly derived from atmospheric processes, their concentrations ought to diminish with depth below the surface. But the relationship between depth and concentration is unknown. Presumably at least some of the oxidant species are

diffusible, for they were present in the soil samples collected from beneath Notch rock at the VL-2 site.

Since the lack of detectable organic compounds within 4–6 cm of the surface seems due to the presence of the oxidants, the likelihood of organic compounds ought to increase with depth. (Organic matter must be present at least transiently on the Martian surface, if from no other source than the infall of carbonaceous chondrites.)

C. The possibility of oases

Although the Martian surface is strikingly similar at two widely separated points when viewed close-up from the two Landers, the surface is strikingly heterogeneous when viewed from orbit. Still, there is no evidence that any of the heterogeneities represent oases that possess characteristics more favorable to terrestrial life than those already enumerated. One dramatic class of heterogeneities, for example, are the channels formed by flowing liquid water. But these channels are too old (probably $\geqslant 10^9$ years or more) to have much bearing on their current suitability for the growth of terrestrial organisms, except that they might possibly contain concentrated deposits of electrolytes and organic compounds.

The orbital infra-red temperature and water vapor measurements also show heterogeneities, but again none of those detected have properties significantly more favorable to terrestrial life than do the larger-scale features. The resolving power of the IRTM is 0.3° (Kieffer *et al.*, 1977), which translates to 8 km at the normal periapsis of 1500 km and 2×8 km for the more recent lowered periapsis of the two Orbiters (300 km). Smaller oases with respect to some of the biologically relevant factors are conceivable (e.g., higher temperatures on south facing slopes in the northern hemisphere; higher temperatures because of heat absorbed by dark objects). It is difficult, however, to conceive of any oasis on the surface of subpolar regions that would be accessible to terrestrial organisms and yet not contain the atmospherically induced oxidants. As mentioned, the subrock samples at VL-2 indicate that some of the oxidants can diffuse in the regolith.

D. Conclusions on the ability of terrestrial organisms to grow or function on Mars

It is thus nearly certain that terrestrial organisms would be incapable of growing on Mars. The low partial pressure of oxygen itself will exclude the functioning of nearly all fungi and all bacteria except strict anaerobes. The temperatures at the surface of subpolar regions often become high enough to be compatible with terrestrial growth. But the possibility of growth and normal function on the surface is rendered essentially nonexistent by the combination of the presence of strong oxidants, the very low probability for the presence of organic compounds of requisite types in required concentrations, the lack of water at activities $\geqslant 0.8$ to 0.9, the strong UV flux, and the nightly decrease in temperature to far below 0 °C. Few terrestrial organisms could function even in the presence of one of these

conditions. We know of none that could function in the presence of all of them simultaneously.

Terrestrial organisms deposited below the surface would be protected from the UV radiation, and they might escape contact with the oxidants. But the protection conferred by increasing depth would be opposed by a rapid drop in the maximum temperature, such that by a depth of 24 cm the temperatures would always be 20 °C or more below the lowest confirmed growth temperature for terrestrial organisms.

At increased depths there is an increased likelihood of encountering ice, the existence of which would enhance the possibility of liquid water. But water that is liquid below −20 °C and is in equilibrium with ice has an activity (a_w) below that which will support the growth of any known terrestrial organisms capable of growing under the partial pressure of oxygen on Mars.

The arguments just presented for subsurface regions generally apply to the residual polar caps as well. As in the subsurface regions, the temperatures mapped by the IRTM are too low to permit the growth of known terrestrial organisms. However, thermal heterogeneities have been detected (Kieffer *et al.*, 1977). The maximum temperatures observed (237 K) are not high enough to permit the growth of Earth organisms, but their presence raises the remote possibility that there exist other undetected heterogeneities where the temperature does rise high enough. But warmer regions will also be drier regions, because the increased vapor pressure associated with higher temperatures would cause water to distill rapidly from these regions and freeze out at the cold trap furnished by the remainder of the residual cap. The water ice itself in the residual caps constitutes a possible source of liquid water, provided that special conditions were present to permit that ice to liquefy rather than to sublime (e.g., freezing point depression by electrolytes). But even then, as in the case of subsurfaces, the temperatures would be too low to permit the growth of terrestrial organisms.

The polar regions would not be immune from the atmospheric oxidants, but chemical interactions between atmosphere and ice might be different from the chemical interactions between atmosphere and regolith.

7. More on the Question of Indigenous Life on Mars

The evidence is that the results of the Viking biology experiments were produced by nonbiological reactions and that terrestrial microorganisms have little, and in most regions of the planet, no probability of growth. But these conclusions do not rule out the possibility that indigenous life forms may currently exist somewhere on Mars or may have existed sometime in the past. The limiting conditions listed in Table VI for terrestrial life are not the limits for conceivable life elsewhere.

There is fairly wide agreement that life, if it exists elsewhere, is based on carbon chemistry and that it requires nitrogen, organic compounds of high information content, energy and substrates to permit the synthesis of the organic compounds, and liquid water. Although, as discussed in Sections 3 and 4, organic compounds

and liquid water have not been detected on Mars, there is no basis for precluding their existence. There is, moreover, strong evidence that liquid water in large quantities existed in the Martian past at least episodically (cf. Section 3).

Nevertheless, the available information on the Martian environment puts severe constraints and presents formidable challenges to any putative Martian organism. A Martian organism that lives on the surface must either be able to protect itself from intense UV radiation and from oxidants apparently powerful enough to decompose organic compounds, or it must synthesize or repair its cellular constituents faster than they are broken down. It must either be able to use water that is so tightly bound as to possess few of the characteristics that make water such a unique solvent for biological systems, or it must expend large amounts of energy to maintain its water in the normal liquid state. A putative Martian organism that lives far enough below the surface to escape the oxidants and the UV will probably also receive insufficient light to photosynthesize.* It will have to derive energy and substrates either heterotrophically or chemoautotrophically. It will face similar problems with respect to water activity as those on the surface plus the added problem of functioning at temperatures that are far below 0 °C. If it exists in thermal equilibrium with its surroundings, the intracellular solute concentrations will have to be multimolar to keep its internal water liquid. If it chooses instead to maintain its contents at temperatures near 0 °C, it will have to expend huge amounts of energy to do so. For example, a spherical Martian organism 2×10^{-4} cm in diameter and encased in efficient insulation $\geqslant 1$ mm thick would have to assimilate and burn about 1000 times its steady state concentration of organic compounds *per second* to maintain its temperature at 0 °C when the surroundings are at −40 °C.

8. Implications from Viking for Past Life on Mars

The Viking data suggest that past conditions on Mars were more conducive to the existence of life. The presence of large quantities of liquid water on the surface means the existence of milder temperatures, it provides a medium for the leaching of essential solutes and for the synthesis of others, and it provides a mechanism for their concentration. But a critical question is how long was the water present on the surface? On Earth, accumulating fossil evidence pushes the date of the origin of terrestrial life earlier and earlier. Thus, the latest evidence shows the presence of microorganisms 3.5 billion years ago (Knoll and Barghoorn, 1977). Consequently, it appears increasingly likely that the origin and establishment of terrestrial life, (once suitable temperatures, pressures, atmospheres, and organic substrates were present) did not require inordinate times. Still, it must have required appreciable time, and the question remains whether the more hospitable epochs in the Martian past had the appropriate characteristics and were of sufficient duration to have permitted the origin and establishment of a Martian biota.

* Although unlikely, the probability is not zero. Sagan and Pollack (1974) have calculated that, although the UV flux is attenuated several million-fold at 0.8 cm below the Martian surface, the flux of visible light would still be 3.8×10^{2} ergs cm^{-2} s^{-1} at that depth.

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