
# Recommendations on Quarantine Policy for Mars, Jupiter, Saturn, Uranus, Neptune, and Titan (1978)

Pages
84

Size
5 x 9

ISBN
0309335981

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

78-0096

0984983 PB83-192070

**Recommendations on Quarantine Policy for Mars, Jupiter, Saturn, Uranus, Neptune, and Titan**

(Final rept)

National Research Council, Washington, DC.

Corp. Source Codes: 019026000

1978 83p

Languages: English

NTIS Prices: PC A05/MF A01 Journal Announcement: GRAI8316

Country of Publication: United States

This report evaluates the current internationally agreed upon planetary quarantine policy, which calls for a probability of contamination (Pc) 0 is less than 0.001 for each planet over the 20 years 1974 to 1994. The probabilistic formulation used by NASA (National Aeronautics and Space Administration) includes Pg, the probability that a terrestrial organism could be deposited on the planet and grown. This report reevaluates the current quarantine policy with respect to Mars, Jupiter, Saturn, Uranus, Neptune, and Titan. The report concludes that the probabilistic basis underlying these quarantine policies is inadequate, and it underemphasizes the experimental search for terrestrial organisms capable of growth under conditions believed to exist on target planets. Alternative experimental approaches are proposed.

Descriptors: *Quarantine; *Planets; Infectious diseases; Contamination; Policies; Foreign countries; Microorganisms; Mars(Planet); Jupiter(Planet); Saturn(Planet); Uranus(Planet); Neptune(Planet); Titan(Planet)

Identifiers: NTISNASNRC

Section Headings: 6E (Biological and Medical Sciences--Clinical Medicine); 22A (Space Technology--Astronautics); 57U (Medicine and Biology--Public Health and Industrial Medicine); 84GE (Space Technology--General)

# Recommendations on Quarantine Policy

## for Mars, Jupiter, Saturn, Uranus, Neptune, and Titan

Committee on Planetary Biology and Chemical Evolution
Space Science Board
Assembly of Mathematical and Physical Sciences



NATIONAL ACADEMY OF SCIENCES
Washington, D.C. 1978

*Available from*

Space Science Board
2101 Constitution Avenue
Washington, D.C. 20418

# Foreword

The planetary missions of the past decade have multiplied our knowledge of the solar system manyfold, and those proposed for the ensuing decade will surely yield equally exciting and provocative results. Scientific prudence requires that during the course of this exploration we guard against perturbing the planets irreversibly and that we take care that ongoing missions not jeopardize future experiments or future discoveries. No discovery would be more important than that of current or past life or life-related organic molecules, and in this area general scientific prudence has been supplemented with a formal international agreement to which the United States is party. Meeting this agreement requires that the probability of contaminating the planets with terrestrial organisms be held below a specified level. The National Aeronautics and Space Administration has the responsibility for determining and taking the steps necessary to meet this policy, but it has requested recommendations from the Space Science Board on the scientific components of the problem. Until three years ago, the Board developed its recommendations through *ad hoc* committees. But since then it has assigned the responsibility for developing quarantine policy to its Committee on Planetary Biology and Chemical Evolution (formerly referred to as the Exobiology Panel). The body of the present report, which constitutes their recommendations for Mars, Jupiter, and Saturn, was reviewed by the Board in its meetings of May 27, 1977, and November 4, 1977, and was adopted unanimously. The report in Appendix C, which constitutes their recommendations for Uranus, Neptune, and Titan, was approved by the Board a year earlier (May 24, 1976) and transmitted to the Administrator, NASA. It is appended here because the Board believes that the basis of its recommendations on quarantine policy for the planets and their satellites should be a matter of public record.

A. G. W. Cameron, *Chairman*
Space Science Board

# Preface

With the launching of Sputnik in 1957, it became inevitable that voyages to other planets would soon follow, voyages that were sure to search for evidence of life or for evidence of organic molecules suggestive of life. Since the scientific impact of such discoveries would be exceedingly high, it was mandatory to take precautions to reduce the chance of their being jeopardized by the contamination of the planets with terrestrial organisms. To coordinate space scientific activities, the International Council of Scientific Unions established a Committee on Space Research (COSPAR), and COSPAR obtained international agreement on a policy on outbound planetary biological contamination control.* The agreement to which the United States is a party is that "The basic probability of one in one thousand ($1 \times 10^{-3}$) that a planet of interest will be contaminated shall be used as a criterion during the period of biological exploration of Mars, Venus, Mercury, Jupiter, or other planets or their satellites that are deemed important for the exploration of life, life precursors, or remnants thereof." The period of exploration has been construed to run from 1974 to 1994 and to consist of 35 landers and 15 flybys. NASA has interpreted these data to mean that, for each mission to each planet, the probability of contamination ($P_c$) should not exceed $10^{-6}$ for landers and $6.4 \times 10^{-5}$ for flybys.

The likelihood of contaminating a planet depends on (1) the number and type of organisms initially on the spacecraft, (2) the fraction that survive the voyage and are released in a viable state onto the planet's surface or into its atmosphere, and (3) the likelihood that the viable organisms find themselves in areas that permit their growth and multiplication. This third component has been assigned the symbol $P_g$. The overall probability of contamination ($P_c$) is the prod-

*We shall refer to this simply as "quarantine."

uct of (1), (2), and (3), and this product must be held sufficiently low to meet COSPAR policy. For landers and planetary probes, little can be done to adjust component (2). The duration of the voyage is fixed by celestial mechanics, and the probability of impacting the planet is we hope close to 1. Orbiters and flybys constitute no quarantine problem as long as they adhere to their expected trajectories and consequently do not impact on the planet. But trajectories that maximize scientific gain usually involve approaches close enough to the planet to raise the possibility that abnormal behavior of the spacecraft will cause it to impact.

For practical purposes, therefore, NASA has considered component (1), the initial microbial burden on the spacecraft, to be the adjustable parameter. And the higher the estimate for $P_g$ the lower is the allowable microbial burden. As the estimated value of $P_g$ rises, increasingly stringent measures must be taken to effect the reduction, measures that can range from sterilization of components to sterilization of the assembled spacecraft or even conceivably to preclusion of the mission.

The irony is that the very pragmatic steps that have to be taken to meet COSPAR requirements are controlled by an elusive and enigmatic quantity–$P_g$, the probability that a terrestrial organism can grow, multiply, and contaminate the target planet. For a planet like Venus, which has surface temperatures that exceed 500°C, $P_g$ is clearly zero. But in the case of Mars and the more distant planets, our ignorance as to the ranges of physical and chemical environments on the planets and our knowledge that terrestrial organisms possess amazing abilities to function in a wide variety of terrestrial environments combine to make the estimation of $P_g$ vexatious and inexact.

It will be evident in reading the Committee's analyses in the body of the report and in Appendix C that another problem in estimating $P_g$ is that the estimates have had to be based largely on epidemiological and probabilistic considerations. As pointed out by Margulis *et al.* (Appendix C and Reference 3), the experimental approach to planetary quarantine has been slighted. There have, for example, been no experiments to determine whether any terrestrial organisms can be found that will grow in aerosol droplets suspended in the high concentrations of hydrogen and methane that prevail in the Jovian atmosphere.

In spite of the difficulties and uncertainties, decisions by NASA on quarantine procedures for planetary missions demand numerical values for $P_g$. Accordingly, the Space Science Board over the past

three years has assigned our Committee the responsibility of making recommendations on $P_g$ for the five planets lying between earth and Pluto. Since other committees had previously estimated $P_g$ for Mars, Jupiter, and Saturn, since the present Voyager mission is to fly by Titan, and since it includes a Uranus option, the first task assigned to us was to estimate $P_g$ for Uranus (and Neptune) and Titan. These estimates, which are given in Appendix C, were approved by the Board and transmitted to the Administrator of NASA in May 1976. The subsequent initiation of work on a Jupiter orbiter with probe (JOP) and the return of major new information on Mars by the highly successful Viking missions led NASA then to request a revaluation of the earlier estimates of $P_g$ for Jupiter, Saturn, and Mars. These revised estimates are given in the body of this report.

Estimations of the probability of growth required us to develop a relationship between knowledge of terrestrial microbial function under terrestrial environmental extremes and knowledge of the physical and chemical conditions found on the target planets. Acquisition of information on the biological aspects was aided by the scientific backgrounds of the members of the Committee. The acquisition of information on the target planets proceeded in two ways. Information on conditions on Mars was gained by the Committee in the course of preparing its recent report *Post-Viking Biological Investigations of Mars.* Information on conditions on the outer planets was obtained chiefly through the participation of two planetary astronomers (A. G. W. Cameron and John Lewis) in an *ad hoc* committee chaired by Lynn Margulis. The relationships between terrestrial biology and planetology that comprise this report were developed during some six meetings of the Committee and invited consultants. Among the latter we wish especially to thank R. Young and N. Horowitz. The Committee also wishes to acknowledge the valuable contributions of its Executive Secretary, Milton W. Rosen, and his staff in organizing our meetings and in the preparation of our report.

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# Contents

# 1
# Introduction

Planetary missions are required to meet the internationally agreed upon COSPAR planetary quarantine policy, which calls for a probability of contamination ($P_c$) of $<1 \times 10^{-3}$ for each planet over the 20 years 1974 to 1994. The probabilistic formulation used by NASA to estimate $P_c$ includes $P_g$, the probability that a terrestrial organism could be deposited on the planet and grow. The current NASA estimate of $P_g$ for Mars required terminal heat sterilization of the Viking Landers and would require comparable sterilization of future Landers. Since the present policy might also limit the scientific return from the Viking Orbiters during the extended Viking mission by putting constraints on the minimum permissible periapsis of these unsterilized crafts, NASA requested that the Space Science Board and its Committee on Planetary Biology and Chemical Evolution re-evaluate the current quarantine policy on Mars in light of the findings from Viking and that it recommend a new policy if appropriate.*

At the same time, a request* was made by NASA for a reassessment of the probability of growth ($P_g$) of terrestrial microorganisms on Jupiter and Saturn. NASA has currently adopted a value of $P_g = 10^{-7}$ based partly on prior recommendations of the Space Science Board. The Board's recommendations were, in turn, based on the probability of the growth of terrestrial microbes on Jupiter and Saturn estimated by an *ad hoc* SSB Committee (R. Goody, N. H. Horowitz, and A. Rich) in 1974. The committee's estimate was derived by assigning a probability of $10^{-1}$ that an organism released into the Jovian or Saturnian atmosphere would be an anaerobe and a proba-

*See Appendix D, letter from Associate Administrator, NASA, to Chairman, SSB, and Appendix E, letter from Chairman, SSB, to Associate Administrator, NASA.

bility of $10^{-6}$ that such an anaerobe would grow. Our more recent analyses of this problem and new data suggest that this earlier estimate of the probability of growth of microorganisms on Jupiter and Saturn is too high. Therefore, a reassessment is appropriate.

In a previous report on *Recommendations on Quarantine Policy for Uranus, Neptune, and Titan*, Appendix C, we stated "that the probabilistic basis underlying COSPAR quarantine policies is inadequate," and that "it underemphasizes the experimental search for terrestrial organisms capable of growth under conditions believed to exist on target planets." Alternative experimental approaches have been proposed (Appendix C and Reference 3), and we recommend that they be pursued. Nevertheless, at present the United States is committed to the probabilistic approach underlying COSPAR policy, and there is as yet no quantitative and operationally useful alternative.

# 2
# Recommendations on Quarantine Policy for Mars Based on the Current Viking Findings

The current NASA policy on the likelihood of growth of terrestrial microorganisms on Mars is based on the December 14, 1970, Space Science Board report, *Review of Sterilization Parameter Probability of Growth ($P_g$).*

The report established the minimum conditions necessary to define a microenvironment on Mars that would support growth of the most "hardy terrestrial organisms." The conditions established were the following:

(a) Water activity ($a_w$) $\geq$0.95.
(b) Temperature $\geq$0° C for at least 0.5 h/day.
(c) Nutrients: At least small amounts of water-soluble nitrogen, sulfur, phosphorus, carbon (and/or light). pH values between 5 and 8.
(d) Attenuation of uv flux by more than $10^3$.
(e) Antinutrients–absence of antimetabolites.

All the above conditions must occur simultaneously, or nearly so.

The report then proceeded to estimate the value of $P_g$, the "estimated probability that growth and spreading of terrestrial organisms on the planet surface will occur." The estimated value of $P_g$ was $3 \times 10^{-9}$, with less than one chance in a thousand that it exceeded $1 \times 10^{-4}$. For the Viking project, NASA adopted a value of $P_g = 10^{-6}$, some three orders of magnitude more favorable to growth than the best estimate of the review committee, but still two orders of magnitude less than the extreme upper limit. The adoption of this

value required terminal heat sterilization of the entire Viking Lander but not of the Orbiter. The value remains NASA policy to date.

## I. VIKING FINDINGS PERTINENT TO QUARANTINE

Estimating the likelihood of the growth of terrestrial organisms on Mars requires a comparison between the known physical and chemical limits to terrestrial growth and the known and inferred conditions present on or just below the Martian surface. Table 1 makes that comparison in abbreviated form. Appendix A discusses in fuller form the inferences that can be drawn from the Viking findings about those physical and chemical characteristics of the Martian surface that are pertinent to the question of the growth of terrestrial microorganisms.

Orbital measurements have covered appreciable fractions of the planet's surface, but the two Landers (VL-1 and VL-2) have sampled only a few square meters of the surface at two 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. (In March 1977 a sample was acquired from a depth of 20 cm, but as of April 1977 an inorganic analysis is the only experiment that has been performed.) Nevertheless, certain extrapolations relevant to the quarantine question can be made with various degrees of confidence to other regions of the planet, to greater depths, and to other seasons of the year.

TABLE 1 Limits for Growth of Terrestrial Organisms

| Factor | 1970 Study | Refs. 2 and 3 | Conditions on Mars[a] |
|---|---|---|---|
| Water activity ($a_w$) | $\geq 0.95$ | $>0.9$ | 0 to 1 |
| Water (liquid) | – | Required | Not detected |
| Temperature | $\geq 0°C$ | $>-15°C$ | +20 to −143°C (see text) |
| pH | 5–8 | $<11.5$ | Not known |
| Ultraviolet radiation[b] | | 0.1 J $cm^{-2}$ | 0.04 J $cm^{-2}$ $min^{-1}$ |
| Ionizing radiation[b] | – | 2–4 Mrad | <500 rad/yr[c] |
| Nutrients | See text and Refs. 2 and 3 | | Organic compounds ≤ppb; most required elements detected (see text)[d] |
| Antimetabolites | None present | | Strong oxidants present (see text)[d] |

[a] Cf. Reference 1; uv flux data from Reference 18.
[b] Limit for survival. Limits for growth are not known.
[c] See p. 11.
[d] At VL-1 and VL-2 sites.

## II. CONCLUSIONS ON THE LIKELIHOOD OF THE GROWTH OF TERRESTRIAL ORGANISMS ON MARS

We turn now to a reassessment of $P_g$, the likelihood of the growth of terrestrial organisms on Mars. We will consider three regions separately: (1) subpolar areas within a few centimeters of the surface, (2) subpolar regions more than a few centimeters below the surface, and (3) the residual polar caps. Finally, we will discuss briefly the likelihood that terrestrial organisms could survive transport at or above the surface from one region to another.

### A. Subpolar Regions within about 6 Centimeters of the Surface

Our conclusion is that no terrestrial organisms could grow within a few centimeters of the surface in the regions lying between the two residual polar caps. We base this judgment on the following of the Viking findings:

– The presence in VL-1 and VL-2 sample of strong oxidants.
– The absence of detectable organic compounds, which (a) attests to the power of the oxidants and (b) renders unlikely the existence of the specific types of organic compounds required for terrestrial heterotrophic organisms.
– The inability of physical shielding by a rock to eliminate the oxidants.

Our conclusion is reinforced by three additional factors that were well known before the mission:

– The unlikelihood of organisms being deposited in regions that receive sufficient visible light to support photosynthetic autotrophy without at the same time receiving lethal fluxes of ultraviolet radiation.*
– The exceedingly low probability for the existence of liquid water with activity ($a_w$) high enough to support terrestrial growth.
– The fact that, even if liquid water were present, vegetative cells would be subjected to daily cycles of injurious freezing; and only vegetative cells can grow.

*Although unlikely, the probability is not zero. Sagan and Pollack[10] have calculated that, although the uv flux is attenuated several millionfold at 0.8 cm below the Martian surface, the flux of visible light would still be $3.8 \times 10^2$ erg $cm^{-2}$ $sec^{-1}$ at that depth.

It is highly likely that the surface conditions enumerated above at the VL-1 and VL-2 sites prevail over the subpolar regions of the planet. This conclusion is based on

1. The similarity in the findings at two widely separated points for the elemental composition of the regolith and for the results of the organic analysis and the gas-exchange experiments.
2. The strong probability that the oxidants are derived from atmospheric reactions or atmosphere–regolith reactions. Accordingly, it is difficult to conceive of regions that would be accessible to terrestrial microorganisms and at the same time be capable of excluding the atmosphere.
3. The fact that the Infrared Thermal Mapper (IRTM) has mapped a sizable fraction of the Martian surface without detecting thermal heterogeneities significantly more favorable to terrestrial growth than those that we have reviewed in Appendix A.

Viking has provided much information that was either not known beforehand or was known only with considerable uncertainty. None of this new information suggests that the Martian surface is less harsh to terrestrial microorganisms than was thought prior to Viking.* On the other hand, two pieces of information indicate that it is harsher than was thought previously: the lack of detectable organic compounds and the presence of strong oxidants even in regions physically shielded from uv.

Our conclusion is that no terrestrial organism could grow under the conditions found by Viking to prevail on subpolar surfaces at the landing sites and none could grow under the conditions that are highly likely to prevail throughout the entire subpolar region. Few if any terrestrial organisms could grow in contact with even one of the adverse conditions cited, much less grow when exposed to all of them simultaneously. Although we cannot absolutely rule out the existence of oases capable of supporting terrestrial life, we believe, for the reasons cited, that the likelihood of their existence is extremely low.

Unfortunately, we know of no quantitative basis for assigning a numerical probability to "extremely low" when no oasis has been

*The demonstration by Viking that the atmosphere contains nitrogen answers an important question that was unknown previously. However, the ignorance prior to Viking of the existence of nitrogen was not a significant factor in prior estimates of the probability of growth of terrestrial organisms.

detected and when the weight of evidence is that none can exist. And yet a numerical value for $P_g$ is required in order to determine what procedures are needed to reduce the microbial burden on future spacecraft to Mars to levels that fulfill current COSPAR quarantine policy. Reluctantly, then, *we recommend for these purposes, and these purposes alone, that NASA adopt a value of* $P_g < 10^{-10}$ *for the subpolar regions of the planet within 6 cm of the surface.** This number, which is more than four orders of magnitude below the current value of $P_g$, reflects the fact that Viking has found the conditions to be considerably harsher to terrestrial life than was heretofore assumed and has obtained evidence that renders the existence of oases far less likely than was heretofore assumed.

## B. Regions More than 6 Centimeters below the Surface of Subpolar Regions

As mentioned, Viking conducted biology experiments and organic analysis on samples obtained from depths of 4–6 cm. Greater depths would be required to reduce or eliminate the lethal surface conditions. The depths required are unknown chiefly because the relation between depth and the presence of oxidants is unknown. However, the maximum temperature falls rapidly with depth. In the northern hemisphere, even at a depth of 4 cm, the maximum temperature is estimated to be 20° below the minimum confirmed growth temperatures (−15°) observed for terrestrial organisms (Appendix B). By a depth of 24 cm, the *maximum* temperature is estimated to be −50°C, some 35° below the minimum confirmed terrestrial growth temperature. In the southern hemisphere, the *maximum* temperature at a depth of 24 cm is estimated to be −35°C, still 20° below the minimum terrestrial growth temperature.[4,8]

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

*We obtain this value by estimating probabilities of $<10^{-2}$ for the presence of liquid water of high enough $a_w$, $<10^{-1}$ for the ability to survive multiple freezing and thawing, $<10^{-1}$ for the avoidance of lethal uv, $\ll 10^{-2}$ for the presence of organic compounds of appropriate types in appropriate concentrations, $\ll 10^{-3}$ for the absence of powerful oxidants, and 0.1 that the deposited microorganism is an anaerobe.

growth of any known terrestrial organism capable of growing under the partial pressure of oxygen on Mars (Appendix B, Figure B.2).[9]

Thus, temperature alone seems an absolute barrier to the growth of any terrestrial organisms at depths below a few tenths of a meter. But again, sufficient uncertainties exist to preclude an absolute statement to this effect; viz.,

– Although the surface temperatures are derived directly from the orbital infrared measurements and are consistent with the direct meteorological measurements at the landing sites 1.5 m above the surface, the estimates of subsurface temperatures require assumptions about the thermal diffusivity of the soil. The range of error is estimated by Kieffer[8] to be 5°C. This error would not be sufficient to change our conclusions, but larger errors are conceivable.

– There could exist heterogeneities below the resolving power of the IRTM (a minimum of 2 km) that have higher temperatures.

– Although there is extensive information on the minimum growth temperatures of terrestrial microorganisms, the remote possibility exists that some unknown organism has a growth minimum below −15°C. We view this as extremely remote because, as indicated in Appendix B, the number of species capable of growth diminishes drastically as the temperature is lowered below 0°C. Furthermore, growth below −15°C is tantamount to growth in ⩾8 osmolal solute, conditions that even at ordinary temperatures preclude the growth of all except halophiles and osmophiles.

– There is the remote possibility that there exists somewhere a narrow zone of subsurface that is deep enough to preclude oxidants and shallow enough to have temperatures high enough to support growth.

Although these uncertainties prevent us from concluding that the possibility for growth is zero, we are still forced to conclude that subsurfaces of Mars are exceedingly harsh for terrestrial life. Accordingly, *for the specific purpose of determining quarantine requirements for future Martian missions, we recommend that NASA adopt a value of* $P_g < 10^{-8}$ *for subsurfaces in the subpolar regions of the planet.*

## C. The Residual Polar Caps

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. 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 for which 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.[4] 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 chemical interactions between atmosphere and regolith.

Our conclusions about the likelihood of growth in the residual polar caps are similar to those reached in Section B above for subsurface subpolar regions—it is extremely low. Nevertheless, because there is more uncertainty about the physical and chemical conditions at the residual polar caps, we believe that these regions should be handled with prudence and *recommend that they be assigned a value of* $P_g < 10^{-7}$.

## D. Transport from Subpolar Regions into the Residual Polar Caps or into Putative Oases

There is little likelihood that any terrestrial organism could survive a voyage on or above the surface requiring more than a few minutes. First, the uv flux on the surface of Mars is $4 \times 10^{-2}$ J cm$^{-2}$ min$^{-1}$, and that flux would kill the most resistant of terrestrial microorganisms in a few minutes (upper terrestrial limit 0.1 J/cm$^2$) (Table 1). Second, organisms protected from the direct exposure to the uv by a layer of soil particles would nevertheless be in contact with the oxidants in those soil particles.

One consequence of these lethal conditions is that our recommended value of $<10^{-7}$ for $P_g$ in the residual polar caps applies only

to terrestrial organisms that are released directly in that region. The $P_g$ for organisms transported into the polar caps from the subpolar regions would be orders of magnitude lower. Similarly, even if Mars were to possess oases that were hospitable to terrestrial life, few if any terrestrial organisms would survive a surface or aerial trip to the oasis and few if any would ever survive an escape from the oasis.

## III. LIMITS TO THE GROWTH OF TERRESTRIAL LIFE VERSUS THE QUESTION OF INDIGENOUS LIFE ON MARS

*The evidence that leads us to the conclusion that terrestrial microorganisms have little and in most regions of the planet no probability of growth does not rule out the possibility that indigenous life forms may exist currently on Mars or may have existed sometime in the past.* The limiting conditions listed in Table 2 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, 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.

It might be argued that, if indigenous life forms do exist, they themselves could constitute micro-oases for the growth of terrestrial organisms. We consider this unlikely. For example, a Martian organism growing in thermal equilibrium with its surroundings at −40°C would be of no value to a terrestrial organism incapable of growing below 0°C. A Martian organism that maintains its temperature at 0°C even when the external temperature is −40°C is conceivable. However, to do so, a spherical organism $2 \times 10^{-4}$ cm in diameter, for example, 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 the 40-degree differential. The problem would be only slightly less serious in a macroscopic Martian organism. Analogous difficulties arise in postulating that the organic compounds in putative Martian biota would be compatible with and utilizable by the enzyme systems of terrestrial microorganisms.

TABLE 2 Estimated Contributions to $P_g$ for Jupiter and Saturn

| Factor | 1974 Jupiter Report | 1976 Uranus Report | Comments |
|---|---|---|---|
| Temperature | 1 | 1 | Assumed between −20 to 100°C |
| Pressure | 1 | 1 | Not a critical parameter for microbiology |
| Radiation | 1 | Not specified but <1 | Deleterious |
| Liquid $H_2O$ | 1 | 1 | Assumed |
| Nutrients | $10^{-1}$ | $<10^{-3}$ [a] | Organics, ions = aqueous solution |
| Anaerobiosis | $10^{-1}$ | $10^{-1}$ | About 0.10 of the earth's microbes are anaerobes, but these are unlikely to be spacecraft contaminants |
| $NH_3$ toxicity | $10^{-2}$ | $<10^{-4}$ [b] | |
| Growth in aerosols | Not specified | $<10^{-3}$ [a] | Completion of life cycle in the atmosphere has never been reported for any earth organisms |
| Convection to lethal temperatures | $10^{-3}$ | $<10^{-3}$ | All models predict that organisms will be carried from water levels to lethal depths; the times required are somewhat model dependent |
| TOTALS | $10^{-7}$ | $<10^{-14}$ | |

[a] Based on more detailed analyses.[2,3]
[b] New information, e.g., Reference 22.

## IV. CONCLUSIONS PERTINENT TO THE CURRENT VIKING ORBITERS

As of August 1977, two years have elapsed since the unsterilized Orbiters were launched from earth. Any organisms on the outer surface of the Orbiter have surely been killed by uv irradiation. Most organisms in the interior of the Orbiter have been subjected to moderate temperatures (10 to 38°C), high vacuum, and some ionizing radiation.[11] Although the cell dehydration associated with the high

vacuum would be lethal to a fraction of the microbial population, many (perhaps 1 to 10 percent) would likely survive.[6,12,13] Some protons from galactic cosmic rays and solar flares would strike organisms in the interior, but the dose would be appreciably less than 500 rad/year,[11,14] and many microorganisms can survive such doses. (The flux of solar protons far exceeds that from galactic source, but the great bulk of the solar protons have energies of $\leq 1$ MeV,[11] and such protons are only capable of penetrating $\leq 0.1$ mm of material with a density of 1, e.g., water.[14]) Conservatively, then, one cannot assume that the microbial burden within the Orbiter has decreased by more than 1 or 2 orders of magnitude since launch.

In spite of the expected survival of a fraction of the original burden of terrestrial microorganisms, our new estimates of the values of $P_g$ lead to the conclusion that COSPAR requirements for planetary quarantine will not be compromised by lowering the periapsis of the Orbiters to 300 km. Indeed, with the new values for $P_g$, still lower periapses for unsterilized Martian orbiters may well be compatible with COSPAR requirements. NASA will probably wish to determine these minimum orbital altitudes before assessing and designing Mars follow-on missions in detail.

## V. QUARANTINE STRATEGY FOR FUTURE MISSIONS TO THE MARTIAN SURFACE

Our Committee has recommended 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 ice–regolith interfaces.[1] Locating these areas and locating sites that are shielded from the powerful atmospheric ultraviolet radiation and the powerful surface oxidants will require subsurface sampling by a soft lander, by penetrators, or by both. 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. The greater the extent to which samples possess these characteristics the greater the priority for the initiation of a second phase of post-Viking biological exploration of Mars—a detailed search for evidence of present or past life on Martian samples returned to earth.

With respect to quarantine considerations for the mission that conducts the first exploratory phase, our estimates for the values of

$P_g$ lead to the conclusion that terminal heat sterilization would not be required in the case of a nominal soft landing in the subpolar regions (Section A) and possibly in other cases as well. However, we would have no objections to sterilization *provided that* it has no impact on the scientific payload of the landers and that it does not increase the mission cost. (We have been informed by representatives of NASA that this may be the case.) Decisions on scientific payloads for the missions should be based on their scientific quality and cost effectiveness. *We would object to the elimination of an experiment or the degradation of its performance because of the imposition of unessential sterilization requirements.*

In the report *Post-Viking Biological Investigations of Mars*,[1] we stated that we consider metabolic-type life-detection experiments on the surface of Mars to be of low priority scientifically. Nevertheless, NASA may decide to include them. If so, a limiting factor with respect to the allowable microbial burden on a soft lander would likely become the avoidance of contaminating the metabolic experiment by terrestrial microorganisms.

# 3
# Revised Recommendations on Quarantine Policy For Jupiter and Saturn: Limits to Growth of Earth Microorganisms on the Outer Planets

In response to a March 1975 request from the Associate Administrator for Space Science, NASA, the Space Science Board asked its Exobiology Panel* to make recommendations on quarantine policy for Uranus, Neptune, and Titan. The Panel's recommendations, which were approved by the Board and transmitted to the Administrator, NASA, on May 28, 1976, are given in Appendix C. The scientific basis for the recommendations was developed by an *ad hoc* committee consisting of Lynn Margulis, Chairman, A. G. W. Cameron, H. O. Halvorson, and John Lewis. Their findings are given in Appendix C. They were also published in expanded form.[3]

The *ad hoc* committee extensively analyzed the chance of growth of terrestrial microorganisms in the atmospheres of the outer planets. Their and our conclusion was that the probability of growth of microorganisms on Uranus and Neptune is nil ($<10^{-14}$). Present ground-based and Pioneer observations indicate that the physical conditions on all four outer planets with respect to parameters affecting biology are probably closely comparable. Thus, the arguments first generated in the context of microbial contamination of Uranus and Neptune apply without essential change to Jupiter and Saturn. *We, therefore, conclude that the probability of growth of earth microorganisms on Jupiter and Saturn is also nil ($<10^{-14}$).*

*Later renamed the Committee on Planetary Biology and Chemical Evolution.

## I. PLANETOLOGICAL CONSIDERATIONS

The analysis was based on the following considerations: It was assumed that growth of all earth organisms requires liquid water within the temperature range from −20 to 100°C. All proposed models of the outer planets predict that liquid water at these temperatures will occur only in the atmospheres. Because it is the most favorable for the growth of earth microorganisms, the model of Weidenschilling and Lewis[21] for atmospheres of the outer planets was used as a basis for our analysis.

## II. COMPARISON OF THE ESTIMATED CONTRIBUTIONS TO $P_g$ IN THE 1974 REPORT WITH THE REVISED ESTIMATES

Table 2 compares the components that contributed to the previous 1974 assessment of $P_g$ for Jupiter and Saturn (see Introduction) with the proposed new value. Both recent data on ammonia toxicity[22] and a more thorough analysis of each of these factors and their interactions lead to this reduction of $P_g$. These new upper limits for $P_g$ are not expected to be sensitive to alterations in the details of planetary models because many outer-planet atmospheric factors are so harsh that microbial life might be excluded on the basis of only one of them. Furthermore, the $P_g$ derived from the values in the right column multiplies out to values that are conservative in that it does not take into account the requirements that organisms must resist simultaneously a large number of limiting factors. It is well known that terrestrial environments combining several adverse factors are effective barriers to growth.

## III. RECOMMENDATIONS

*As shown in Table 2 we estimate* $P_g$ *for the outer planets, Jupiter, Saturn, Uranus, and Neptune, to be* $<10^{-14}$. When this low value is inserted into the probabilistic formulation used by NASA, it leads to the conclusion that sterlization of spacecraft and their payloads is not required to meet the COSPAR requirements described in the Introduction. Nevertheless, we continue to support our prior recommendation (Appendix C) and that of COSPAR[23] that spacecraft and payloads bound for Jupiter and Saturn be assembled under cleanroom conditions comparable with the class 100,000 cleanroom procedures used for the current Viking. (A document detailing these procedures is currently in preparation.[24]) One reason for cleanroom assembly is to reduce the possibility that the growth of microorganisms in the

spacecraft might compromise the function of the spacecraft or its scientific payload (Appendix C).

Except for Titan, the Committee has not considered the satellites of the outer planets. For Titan, the Committee recommended a tentative value of $P_g$ of $10^{-10}$ (Appendix C).

Virtually all models of the Jovian planets agree that hydrogen gas is a predominant constituent of the atmospheres. Our estimate of $P_g$ has not included possible effects of high hydrogen concentrations on the growth of potential contaminant organisms. Careful studies designed to select terrestrial microbes capable of growth in high pressures of H (and $CH_4$) are lacking. We suggest that these experiments would not only add valuable information to basic microbiology but might provide results immediately applicable to planetary quarantine as well.

# Appendix A: Findings from Viking Pertinent to the Possible Growth of Terrestrial Microorganisms on Mars*

## I. DEFINITIVE FINDINGS FROM VIKING

### A. Water

The gas chromatograph–mass spectrometer (GCMS) has detected less than 0.1 percent water in soil samples (several tenths of a percent in one sample collected from beneath a rock). The current belief is that this water represents mineral hydrate water of moderate or low thermal stability. Neither this instrument nor the others on the lander were designed to detect free liquid water, nor have they done so.

Unfortunately, Viking carried no instrument to measure relative humidity. However, indirect evidence (e.g., cloud formation) indicates that saturation does occur in the atmosphere.

The probability for the existence of liquid water anywhere on the planet remains low. The surface temperatures and atmospheric pressures preclude the existence of pure bulk liquid water under equilibrium conditions. However, there continue to be three remote possibilities for the existence of liquid water: (1) 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 heat), and (3) water that has its chemical potential (and hence freezing point) lowered by the presence of dissolved solutes. The solutes could be one or more of the several salts that are almost certainly present. The eutectic points of

*Pertinent references not cited here will be found in Reference 1.

salts like $CaCl_2$ $MgCl_2$, and $K_2CO_3$ are below −30°C; hence their presence would permit stable liquid water down to these temperatures. The electrolyte concentrations, however, would be multimolar.

Another argument against the existence of liquid water at the landing sites is the findings of the Labelled Release and Gas Exchange biology experiments. In both cases, the initial addition of water vapor or liquid water to the soil samples dissipated the reactants so that further additions produced no further reactions (release of $^{14}CO_2$ 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.

## B. Temperature

The maximum temperatures observed at the surface of the landing sites during the summer–autumn observation period were −2 to −3°C. This is below the minimum growth temperature of most terrestrial microorganisms, although, as discussed later and in Appendix B, 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.[4]

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.[5-7] And any terrestrial microorganism that is to grow on Mars must by definition be in the vegetative state to carry out such growth.

## C. Lack of Detected 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 were undetectable or detectable only at reduced sensitivities.

The inability to detect organic compounds does not, of course, prove that none was present. But even if trace amounts of organic

compounds are in fact present in the soil of the landing sites, the probability is remote that these would provide a nutrient medium that could be used by terrestrial microorganisms (see Reference 3 for further discussion).

## D. Elemental

The biologically vital element nitrogen has now been shown to be in the Martian atmosphere. Calcium, sulfur, magnesium, chlorine, and probably potassium and phosphorus have also been detected in soil samples. All six are essential to terrestrial living systems. Instrument limitations precluded the detection of sodium, but there is no reason to believe that it is not present, although probably only in low concentrations.

One striking finding is that the elemental composition of the samples was nearly identical at the two widely separated landing sites.[26] 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.[27]

## E. Oxidants

Two lines of evidence indicate that strong oxidants are present in at least the top few centimeters of the regolith at the landing sites. The first line of evidence comes from orbital measurements of the atmosphere and from modeling. One model predicts the existence of active strongly oxidizing species, especially hydrogen peroxide. Second, the gas-exchange experiment (GEX) on the Viking landers showed the release of up to nearly a micromole of oxygen when samples were humidified with water and warmed to -10°C.

The GEX experiment suggests that oxidants are present to at least the 4–6-cm depth from which samples were acquired. The experiment also showed that the oxidants were present in samples collected from beneath a rock, a rock that presumably had laid undisturbed for many years. Finally, the experiment showed that the oxidants were present at both landing sites.[28]

The oxidants are believed to be responsible for the lack of detectable organic compounds, i.e., they have decomposed them.

## II. 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 Martian winter will drop far lower than those experienced during the Lander experiments. Estimates from the infrared thermal mapper (IRTM) indicate that the *maximum* surface temperature will fall below −15°C (the minimal terrestrial growth temperature—see Appendix B) 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.[8] (Orbital IRTM measurements during winter will become available during the ensuing months.)

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 degrees in longitude, the results of the gas-exchange (GEX) and labeled release (LR) biology experiments and of the organic and inorganic analyses at the two sites were either similar or essentially identical.* Strong similarities were evident as well from the imaging experiment and from the atmospheric analyses. As noted, 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 extrapolation can also be made to depths below the 4–6 cm sampled by the Landers.

### A. Water

Several Viking experiments have confirmed or strengthened the inference that large amounts of water are locked beneath the surface in the form of ice. Subsurface liquid water is conceivable; however,

*Samples from the two sites in the Pyrolytic Release (PR) experiment, however, responded differently to the addition of water vapor.[25] The experimenters 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.

because of the low temperatures at subsurfaces (see below), the existence of liquid water in an equilibrium state would require multimolar concentrations of electrolytes (see Appendix B, Table B.1).

## B. Temperature

The maximum summer temperatures some 6 cm below the surface at the VL-1 and VL-2 sites are estimated from the IRTM measurements to be −35°C.[8] This temperature is 20° *below* the minimum confirmed growth temperature for terrestrial microorganisms. 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 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° warmer than at the present landing sites. 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° below the minimum confirmed terrestrial growth temperature.[4]

## C. Ultraviolet Light

As shown in Table 1, the flux of ultraviolet radiation impinging on the Martian surface would be rapidly lethal to any terrestrial organism. However, the uv flux is sharply attenuated below the surface. For example, Sagan and Pollack[10] estimate an attenuation of several millionfold at a depth of 0.8 cm.

## D. 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 diffusable, for they were present in the soil samples collected from beneath the 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.)

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, is the huge channels that were almost certainly formed by flowing liquid water. But these channels are too old (probably $\geq 10^9$ years) 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 infrared 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°, which translates to 8 km at the normal periapsis of 1500 km and 1.6 km for the now lowered periapsis of Orbiter 1 (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 sample at VL-2 indicates that some of the oxidants can diffuse in the regolith.

# Appendix B
# Minimum Temperature for Terrestrial Microbial Growth

The most thorough review known to us of the minimum growth temperatures of terrestrial microorganisms is that of Michener and Elliott.[15] A histogram summarizing their findings on reports of growth below 0°C is shown in Figure B.1. Many of these reports are based on incubation times of over a year. We separate bacteria from fungi because the latter are nearly all aerobic and would be incapable of growing at Martian partial pressures of oxygen. 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 confirmed. Michener and Elliott point out that "The best evidence that growth does not generally occur in foods in this temperature 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 study by Fennema *et al.*[16] confirms Michener and Elliott's conclusion that microbial growth in foods does not occur at −18°C.

This inability of organisms to grow below about −15°C is consistent with the known physical state of aqueous solutions at these temperatures. As Table B.1 shows, when solutions of sodium chloride in water, for example, are equilibrated at various subzero temperatures, the concentrations in the unfrozen portions exceed 4 molal below −15°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 $\phi$ is the osmotic coefficient, $\nu$ the number of species into which the solute dissociates, and $m$ is the molality.[17] Aside from the toxic effects to nearly all microorganisms of such high concentrations of electrolytes, the high concentrations also depress the water

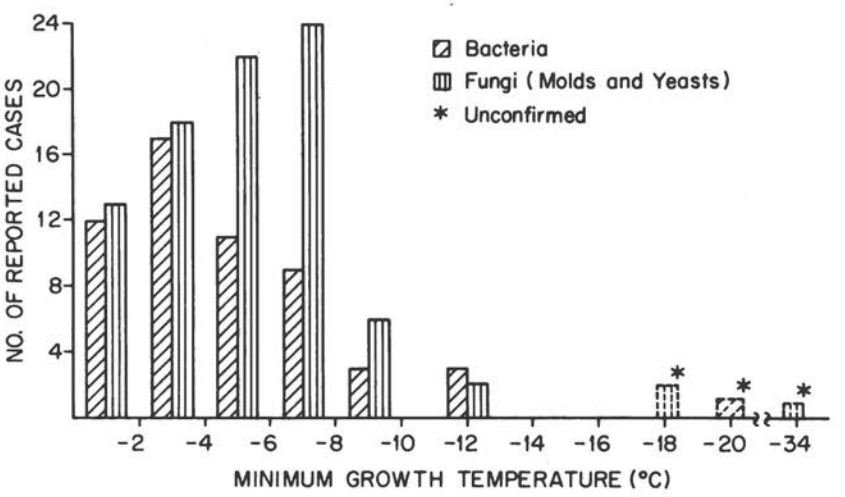


FIGURE B.1 Reported cases of microbial growth below 0°C. (Adapted from Reference 15.)

TABLE B.1 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 |
| -15 | 3.96 | 0.86 |
| -16 | 4.17 | 0.85 |
| -18 | 4.58 | 0.84 |
| -20 | 5.00 | 0.82 |

[a] From Reference 19.

[b] $a_w = p_{H_2O}$ (solution)/$p_{H_2O}$ (liquid, pure)

$\equiv p_{ice}/p_{H_2O}$ (liquid pure).

Calculated from data in Reference 20. See Figure B.2.

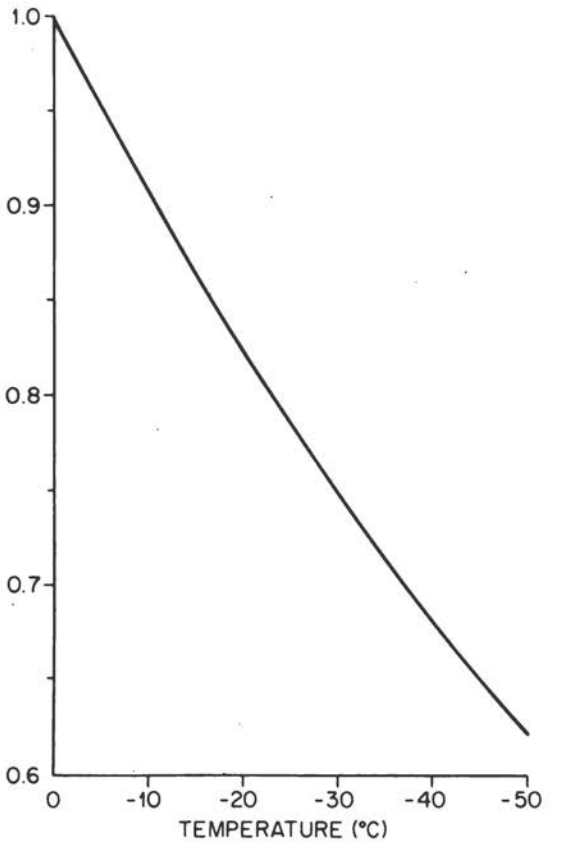


FIGURE B.2 Water activity $a_w = p_{ice}/p_{H_2O}$ of a solution in equilibrium with ice as a function of temperature.

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 B-1 and Figure B-2, the values of $a_w$ at −14, −16, −18, and −20°C are 0.87, 0.85, 0.84, and 0.82, respectively.

# Appendix C
# Recommendations on Quarantine Policy for Uranus, Neptune, and Titan

Exobiology Panel
Space Science Board
May 24, 1976

Peter Mazur, Oak Ridge National Laboratory, *Chairman*
Elso S. Barghoorn, Harvard University
Charles D. Cox, University of Massachusetts
Harlyn O. Halvorson, Brandeis University
Thomas H. Jukes, University of California, Berkeley
Isaac R. Kaplan, University of California, Los Angeles
Lynn Margulis, Boston University

In response to a request of March 22, 1975 (Attachment C.1) from Noel Hinners, Associate Administrator for Space Science, NASA, the Space Science Board asked this Panel to make recommendations to the Board on quarantine policy for spacecraft flights to Uranus and Neptune. The Panel in turn established an interdisciplinary *ad hoc* Committee to analyze the problem and provide recommendations and supporting documentation. The Committee consisted of two members of the Panel (Lynn Margulis and H. O. Halvorson) and two others (A. G. W. Cameron, Harvard University, and John Lewis, MIT). It was chaired by Dr. Margulis.

An intermediate draft of their report was discussed by the Panel at its September 1975 meeting. The draft was also transmitted to some 25 biologists, atmospheric physicists, and astronomers for critique (Attachment C.2). After being revised on the basis of these comments, the Committee's report was discussed in detail at the Panel's March 11-12, 1976, meeting. After further revision, it has been accepted by the Panel and is attached (Attachment C.3).

While the Panel concurs in general with the analysis, documentation, and conclusions of the Committee, the Panel's specific recommendations to the Board are those stated here.

## I. RECOMMENDATIONS ON PLANETARY QUARANTINE

We agree with the Committee that the probabilistic basis underlying COSPAR planetary quarantine policies is inadequate (see Section II.A of Attachment C.3), but the United States is as of now committed to this internationally agreed upon policy, and there is as yet no quantitative and operationally useful alternative.

The policy calls for a probability of contamination ($P_c$) $< 1 \times 10^{-3}$ for each planet over the 20 years 1974 to 1994. It makes the assumption that there will be 35 landers and 15 flybys during this period. The probabilistic formulation used to estimate $P_c$ is given in Section II of the Committee's report, and it includes $P_{(g)}$, the probability of growth.

1. *We conclude that for flybys and probes of Uranus and Neptune, $P_{(g)}$ is sufficiently low so that $P_c$ will be less than the COSPAR requirement even if no special precautions are taken to reduce $m_i(0)$ the initial microbial burden at launch.*

*Nevertheless, for reasons unrelated to the calculation of $P_{(g)}$ and $P_c$, we recommend that spacecraft bound for these two planets be assembled under "clean-room" conditions comparable with those in*

*the Viking program (but without the sterilization procedures used on Viking).*

2. The *ad hoc* Committee's report and the Panel's recommendations address only the question of the contamination of Uranus, Neptune, and Titan by terrestrial organisms. They do not address, and did not consider, the separate question of the possibility of indigenous life on the outer planets. Our conclusion that terrestrial organisms have nil probability of growth does not, of course, rule out the possibility of indigenous life forms. Nor does it alter the fact that an understanding of the organic chemistry of the atmospheres of the outer planets will be of great exobiological interest. It is for this latter reason, in part, that we recommend "clean-room" assembly of spacecraft to reduce the possibility of introducing undesirable quantities of organic contaminants.

3. *Titan.* The predecessors to this Panel stated in the 1974 report* "In view of the fact that current knowledge of the surface and atmosphere of Titan is very fragmentary and that new evidence is accumulating rapidly, we believe that Titan should be treated circumspectly. We therefore *recommend* that, pending further information, $P_{(g)} = 0.1$ be adopted for Titan, this representing our belief that at best only anaerobes could live in that satellite's environment."

Although the present Panel and its *ad hoc* Committee were not charged with preparing recommendations on Titan, the accumulation of new information made it desirable to do so. The Committee has analyzed Titan models in detail and concludes that a model not totally excluding the possibility of the growth of terrestrial microorganisms is barely conceivable. Their estimate of $P_{(g)}$ on the basis of this improbably optimistic model is $10^{-10}$. We recommend the adoption of this value until data on Titan are available from MJS.

## II. TECHNICAL COMMENTS

### A. Choice of Model

We concur with the Committee's view that the presence of liquid water and temperatures between 253 and 373 K are mandatory for the growth of any terrestrial organisms. They conclude that these

**Quarantine Considerations for Jupiter and Saturn Missions*, NAS-NRC, Washington, D.C., 1971, revised 1974.

conditions are fulfilled in the published model of Weidenschilling and Lewis. No respondent has challenged that conclusion.

Three consequences follow from this model:

1. Terrestrial biology could function nowhere but in the atmosphere.
2. Regions of the atmosphere that are biologically acceptable in terms of temperature and liquid water will contain $NH_3$ at concentrations of 0.3 to 7 M.
3. The atmosphere is convective, and organisms will be convected out of the "hospitable" regions of the atmosphere into regions with temperatures high enough to produce complete sterilization.

## B. Planetary Atmospheres as a Barrier to Microbial Growth

The earth's microbial population is very large, possibly on the order of $10^{25}$ organisms ($\sim 10^4/cm^3 \times 10^{21}$ $cm^3$) in the top two meters of the soil. Representatives of this population occupy and function in an extraordinary diversity of environments including niches with high and low temperatures, high and low pH's, and high salinity or pressure; and they function with a wide variety of substrates. In view of the large population and its demonstrated ecological versatility, it seems rather extraordinary that there is no evidence for the functional occupancy by microbes of one vast niche–the earth's atmosphere. Biological aerosols have been studied extensively in the laboratory, and Dimmick and Chatigny have recently pointed out that "Not a single instance has been reported where the biological decay [rate of disappearance of viable cells in aerosols] was less than the physical decay [physical fallout], a situation that would have to exist if true propagation has occurred." (However, Dimmick and his colleagues have recently published evidence consistent with the existence of possible limited and transient metabolic activity by an aerobic bacterium in aerosols formed under optimal conditions which included the presence of appropriate nutrients; on the other hand, no activity has been detected with anaerobic cells.*)

*R. L. Dimmick and M. A. Chatigny, Possibility of growth of airborne microbes in outer planetary atmospheres (preprint); R. L. Dimmick, Patricia A. Straat, H. Wolochow, G. V. Levin, M. A. Chatigny, and J. R. Schrot, Evidence for metabolic activity of airborne bacteria, *J. Aerosol Sci. 6*, 1975 (in press); Letter, R. L. Dimmick to P. Mazur, 1/27/76.

Upon reflection, the apparent inability of the terrestrial atmosphere to support a functional microbial population is not so surprising. The chief barriers are probably two: water and nutrients. The great preponderance of microorganisms grow (and presumably have evolved) in dilute aqueous solutions in which the osmolality* is $\leqslant 1.1$ and the water activity ($a_w$)† is therefore $\geqslant 0.98$. Relative humidities $\geqslant 98$ percent in the earth's atmosphere are transient, and, therefore, water droplets with $a_w \geqslant 0.98$ exist only transiently. If solutes are present in atmospheric water droplets, a drop in relative humidity (RH) to, say, 95 percent will raise the equilibrium osmolality in aqueous solutions to 3 osmolal, and concentrations of this magnitude commonly induce cell stasis or death. Furthermore, the nonvolatility of most biological solutes and nutrients will produce solute compositions in droplets that will in all likelihood be qualitatively and quantitatively different from those experienced by organisms on the earth's surface. Alternatively, if solutes were essentially absent in an atmospheric water droplet, a drop in RH below 100 percent would lead to the vaporization of all liquid water that has not had its activity reduced by being bound by electrostatic or capillary forces to solid surfaces. The altered solute composition or the loss of all free water would be likely to produce stasis or death in large fractions of the terrestrial microbial population.

The apparent inability of terrestrial microorganisms to reproduce in the atmosphere exists in spite of the fact that the proper nutrients to support growth exist on earth and in spite of the fact that organisms in the atmosphere are not usually in contact with high concentrations of gases like ammonia and are not subject to convection into regions exhibiting pyrolytic temperatures. In the Uranian and Neptunian atmospheres, as the Committee report shows, the existence of proper nutrients is unlikely, and the existence of high concentrations of ammonia and convection to pyrolytic temperatures are likely. These factors do not enhance the probability of growth of any terrestrial organism deposited in their atmospheres.

*Osmolality is defined as $\phi\nu m$, where $\phi$ is the osmotic coefficient, $\nu$ the number of species into which the solute dissociates, and $m$ is the molality.

†Water activity ($a_w$) is numerically equal to relative humidity expressed as a fraction and is equal to $p/p_0$, where $p$ is the vapor pressure of waters in an aqueous solution and $p_0$ is the vapor pressure of pure water at the same temperature. The relation between $a_w$ and osmolality is $\ln a_w = -0.018\ \phi\nu m$.

## C. Definition of "Nil"

The Committee concludes that $P_{(g)}$ for Uranus and Neptune is "Nil." The formulas used by NASA for the calculation of $P_C$ require a numerical value of $P_{(g)}$, and for this purpose (and this alone) we suggest defining "Nil" as $<10^{-14}$. We obtain this value by assigning probabilities of $<10^{-3}$ for growth in an atmosphere, $<10^{-4}$ for the ability to grow in contact with ammonia, $<10^{-3}$ for the ability to survive convection, $<10^{-3}$ for the presence of the qualitatively and quantitatively appropriate concentrations of nutrients, and $10^{-1}$ for precluding all but anaerobic organisms.

## D. Models versus Reality

A number of respondents raised the question of one's confidence in the correctness of the models that provided the bases for calculating $P_{(g)}$'s. They doubted whether that confidence would exceed 0.99, much less $(1-10^{-14})$. In our view, that is not the pertinent question. The pertinent question is what is the probability that reality on Uranus, Neptune, and Titan is far more hospitable to the growth of terrestrial organisms than the conditions predicted by the selected models? Although a definitive answer is impossible with present knowledge, we make the following points:

1. The models were not chosen because they are the most likely, but, because of the quantitative models proposed, they are the *most* favorable to terrestrial life. The others lead to temperature and states of water that totally exclude terrestrial growth.
2. Acceptance of the Weidenschilling and Lewis model for Uranus and Neptune results in the atmosphere being convective, an atmospheric location for the biologically favorable zone, and the presence of high concentrations of ammonia in that zone. These conditions are not selected arbitrarily but are mandated by a combination of observational data and physical and physical-chemical principles.
3. The conditions, both taken singly and in combination, are so harsh to the growth of terrestrial life that the "nil" value for $P_{(g)}$ is not sensitive to the precise numerical environmental values derived from the model. Indeed when ranges of numerical values are possible, the Committee generally selected those more favorable to terrestrial life (e.g., longer mixing times; lower concentrates of ammonia).

4. Similarly, the conditions are so harsh that the conclusion about $P_{(g)}$ being "nil" is not sensitive to the discovery of some rare exceptionally hardy organism as, for example, the recent discovery of an anaerobic halophile. The flexibility of terrestrial germ plasm is remarkable in permitting organisms to occupy incredibly diverse environments, but we must emphasize that organisms pay a price for the ability to survive and function in a given environmental extreme; namely, they become more demanding with respect to the tolerable limits of other environmental factors. Thus, spores *survive* many harsh environments, but in the sporulated state they are incapable of growth; halophiles grow in high sodium chloride concentrations, but nearly all require oxygen and are rapidly lysed at "normal" levels of sodium chloride; psychrophiles will grow at lower temperatures than mesophiles but are often more sensitive to freezing. The result is that environments that impose combinations of adverse environments can be highly effective barriers to terrestrial growth.

The Panel is able to proceed no further on the basis of scientific analysis. We do not know how to assign a probability to the future emergence of models of Uranus, Neptune, or Titan that are more favorable to terrestrial life. We do not know how to assign a probability to the existence on the outer planets of niches that are more favorable to the growth of terrestrial organisms. Knowing no way to assign probabilities to these factors, we know of no scientific basis for assigning a probabilistic "safety factor" or "insurance" to the Committee's value of $P_{(g)}$. Logically, the only procedure to ensure safety would be to assume $P_{(g)} = 1$. Although logically defensible, we would find such a procedure indefensible on a cost/benefit basis. A likely cost would be cancellation of the missions.

## III. REDUCTION OF BIOLOAD ON SPACECRAFT

As mentioned, we concur with the Committee's conclusion that $P_{(g)}$ is "nil" for Uranus and Neptune and $10^{-10}$ for Titan. Acceptance of these values leads to the conclusion that no precautions to reduce the microbial burden on spacecraft need to be taken for the purpose of meeting the COSPAR agreement on $P_c$.

Although indeterminable, there exists some true value of $P_{(g)}$ for each outer planet. If this value is truly greater than zero, then reducing the microbial burden on spacecraft at launch will produce a concomitant reduction in the true value of $P_c$. A spectrum of procedures is available for reducing the bioload to any desired extent.

The dollar cost is a power function of effectiveness. Increasingly effective sterilization procedures have other costs as well: time delays, restrictions on materials used in scientific payloads, possible adverse effects on mission reliability, and possible constraints on the scientific experiments. Decisions on the desirability of incurring one or more of these costs cannot be justified on the basis of scientific analysis but fall rather in the realm of what Weinberg terms "trans-science." To repeat, our *scientific* judgment is that no sterilization procedures are required to meet the COSPAR agreement on $P_C$.

There are, however, arguments unrelated to $P_{(g)}$ or $P_C$ in favor of taking one step to reduce the microbial burden in spacecraft, namely, clean-room assembly. One biological argument is that in the absence of clean-room assembly the probability is higher than "nil" of introducing clumps of soil and other organic materials into the spacecraft. Such clumps could provide moisture and nutrients for limited growth of microorganisms, especially fungi, during the flight. The growth of organisms could compromise the function of the spacecraft or of the scientific payload. Fungi, for example, are notorious for growing on and even etching optical surfaces. In addition, the introduction of adventitious organic material into planetary atmospheres may *per se* be undesirable.

NASA has informed us (Attachment C.4) that based on experience with Mariners the cost of clean-room assembly is about 0.2 percent of project costs. We consider this an acceptably low cost/benefit ratio.

## IV. ALTERNATIVES TO CURRENT STRATEGY FOR ESTABLISHING QUARANTINE POLICY– AN EXPERIMENTAL APPROACH

The *ad hoc* Committee correctly points out that current quarantine strategy emphasizes the assigning of probabilities to qualitatively and quantitatively unknown phenomena and underemphasizes the experimental search for terrestrial organisms capable of growth under conditions believed to exist on target planets. For example, in the case of Uranus, it should be feasible to search experimentally for organisms that are capable of surviving several months under conditions existing inside the spacecraft and are then capable of growth while suspended in atmospheres lacking oxygen and containing ammonia. As the Committee suggests, other parameters, the values of which are unknown or uncertain on the planets, could be optimized to pro-

mote growth. While such a procedure could never eliminate the possibility that the "right" organism exists but is not detected, increasing numbers of negative experiments would make that possibility increasingly remote. Positive results on the other hand would not only identify microbes of potential quarantine concern but would make available species of inherent scientific interest. Limited approaches of this sort were undertaken for Mars and, we understand, are under way for Jupiter.

Viking has been sterilized and heads for Mars. Data will, we hope, return this fall. Quarantine policy for the 1977 MJS (possible MJSU) will of necessity be determined by the existing strategy. Venus and Mercury offer no quarantine problem. This Panel *recommends*, therefore, that decisions as to when, or whether, to fully implement the Committee's suggested alternative strategy await the return of data from these committed flights. On the other hand, the areas of research suggested by the Committee in Section VI of its report are of considerable interest to microbial ecology, apart from their relation to questions of planetary quarantine.

## Attachment C.1

Mr. Milton Rosen
Executive Secretary, Space Science Board
National Academy of Sciences
2101 Constitution Avenue, NW
Washington, DC 20418

Dear Mr. Rosen:

You will recall that you recently submitted to us the recommendations of the Space Science Board on the caution that should be exercised to protect Jupiter and Saturn from terrestrial biological contamination. This same report demurred on similar recommendations for Uranus and Neptune for the reason that not enough new knowledge on these planets was forthcoming to update the old model.

We quite understand this position, but in view of active discussion of possible Uranus atmospheric entry probes to be launched during the 1979–84 period, our planners need the best possible advice on the degree of caution, if any, that should be observed in studying that planet. Similar consideration of Neptune would also be most useful. Past experience has demonstrated that such recommendations must be the result of a dynamic process with early evaluations being modified as we progressively learn more about each of the planets.

We request, therefore, that the Space Science Board review the available knowledge on Uranus and Neptune and give us recommendations, prior to the COSPAR meeting in June 1975 if possible, for planetary quarantine policy toward the first Uranus and Neptune entry probes. We anticipate, of course, that any recommendations will be provisional, to be revised as additional knowledge becomes available.

Thank you for your assistance.

Sincerely yours,

NOEL W. HINNERS
Associate Administrator for Space Science
National Aeronautics and Space Administration
Washington, D.C. 20546

## Attachment C.2

**Individuals Asked to Comment on Draft of the *ad hoc* Committee's Report**

Martin Alexander, Cornell University
Thomas Brock, Indiana University
C. S. Cox, Porton, England
Robert Danielson, Princeton University
Leo Daspit, Langley Research Center
Paul Deal, Ames Research Center
R. L. Dimmick, University of California, Berkeley
Harry Eagle, Yeshiva University
Richard Goody, Harvard University
Norman Horowitz, California Institute of Technology
Andrew Ingersoll, California Institute of Technology
Holger Jannasch, Woods Hole Oceanographic Institution
Harold Klein, Ames Research Center
Joshua J. Lederberg, Stanford University
Conway Leovy, Washington University
G. Levin, Biospherics Research Inc.
Elliot Levinthal, Stanford University
A. G. Marr, University of California, Davis
J. S. Martin, Ames Research Center
Richard Morita, Oregon State University
Tobias Owen, State University of New York, Stony Brook
Vance Oyama, Ames Research Center
Cyril Ponnamperuma, University of Maryland

S. I. Rasool, National Aeronautics and Space Administration
S. C. Rittenberg, University of California, Los Angeles
Carl Sagan, Cornell University
Sanford Siegel, University of Hawaii
G. A. Soffen, Jet Propulsion Laboratory
R. Stanier, University of California, Berkeley
Ralph Wolff, University of Illinois
Richard Young, National Aeronautics and Space Administration

## Attachment C.3

**ON CONTAMINATION OF THE OUTER PLANETS BY EARTH ORGANISMS**

National Academy of Sciences
Panel on Exobiology Report of the
Ad Hoc Committee on Biological Contamination
Of
Outer Planets and Satellites

March 20, 1976

Submitted to: Peter Mazur, Chairman, Exobiology Panel
From: H. O. Halvorson (Brandeis), J. Lewis (MIT), A. G. W. Cameron (Harvard), L. Margulis (Boston University, Chairman). Consultants: Ronald Prinn, Peter Stone (Department of Meteorology, MIT)

This document contains:



## INTRODUCTION

In response to a request of March 1975 from Noel Hinners, Associate Administrator for Space Science, NASA, the Space Science Board

asked its Exobiology Panel to make recommendations to the Board on quarantine policies for spacecraft flights to Uranus and Neptune. The Panel in turn established an interdisciplinary *ad hoc* Committee to analyze the problem and provide recommendations and supporting documents. The Committee consisted of two members of the Panel (Lynn Margulis and H. O. Halvorson) and two other (A. G. W. Cameron and John Lewis); it was chaired by Dr. Margulis.

A prior report of the Exobiology Panel,* dealing with Jupiter and Saturn, also commented on the Saturnian satellite Titan in these words, "In view of the fact that current knowledge of the surface and atmosphere of Titan is very fragmentary and that new evidence is accumulating rapidly, we believe that Titan should be treated circumspectly. We therefore recommend that, pending further information, $P_{(g)} = 0.1$ be adopted for Titan."

Although the present Panel and its *ad hoc* Committee were not charged with preparing recommendations on Titan, the accumulation of new information made it desirable to do so. The Committee has analyzed Titan models in detail and concludes that a model not totally excluding the possibility of the growth of terrestrial microorganisms is barely conceivable. The estimate of $P_{(g)}$ on the basis of this model is $10^{-10}$.

In its prior report,* the Panel brought into question the probabilistic approach to evaluating the risk of planetary contamination by noting that the spread between the minimum and maximum values of $P_c$ was $10^8$, i.e., eight orders of magnitude. Their report stated, ". . . the spread between maximum and minimum figures confirms the view expressed by one consultant that the numbers game is essentially meaningless. We believe, however, that our exercise has been worthwhile to the extent that it exposes no obvious conflict between our intentions and our international obligations. We therefore conclude that our international obligations with respect to outer planet missions are fully satisfied by means of quarantine procedures which do not impair the capability or significantly increase the cost of the missions, i.e., surface sterilization of entry probes, but only normal clean room assembly of other spacecraft."

The present Committee, therefore, felt it was incumbent upon them not only to evaluate the probability of contaminating Uranus and Neptune, but also to consider the merits of the probabilistic approach.

**Quarantine Considerations for Jupiter and Saturn Missions*, NAS-NRC, Washington, D.C., 1971, revised 1974.

## I. ABSTRACT OF THE CONTENTS OF THIS DOCUMENT

1. We advocate the abandonment, in principle, of the probabilistic approach to the estimation of growth of earth organisms on spacecraft, planets, and satellites in the solar system. We do not support an approach that estimates probabilities of qualitatively unknown phenomena.

2. We suggest the formation of a new interdisciplinary committee (to design a replacement for the current probabilistic approach) based on a different strategy. We recommend a strategy that involves identification and intensive study of those organisms most likely to thrive under known conditions for each of the planets respectively. (Unknown environmental conditions may be allowed to vary optimally.)

3. We have examined various models for the outer planets and chosen extreme ones from the point of view of the presence of liquid water in reasonable temperature ranges (0 to 100°C). We have concluded that no presently known organism could multiply in the atmospheres of the outer planets Uranus and Neptune, even on the basis of the most earth-like models.

4. Titan, the $CH_4$-rich moon of Saturn, may be more hospitable for terrestrial organisms than any of the other objects of the outer solar system. We estimate the probability of growth of an earth organism in the atmosphere of Titan about $10^{-10}$. We recognize that revision of our views concerning Titan must occur as more is learned about this satellite.

5. Further specific terrestrial experimentation is recommended.

## II. RECOMMENDATIONS ON GENERAL POLICY CONCERNING PLANETARY QUARANTINE

We recognize that decisions on quarantine policy affect many people and their activities. In the past these decisions have been based on the following formulation:

$$P_c = m_i(0) \cdot P_{(vt)} \cdot P_{(uv)} \cdot P_{(a)} \cdot P_{(sa)} \cdot P_{(r)} \cdot P_{(g)} \text{ where}$$

| | |
|---|---|
| $P_c$ | Probability of contamination |
| $m_i(0)$ | Initial microbial burden (at launch, after decontamination) |
| $P_{(vt)}$ | Probability of surviving space vacuum-temperature |
| $P_{(uv)}$ | Probability of surviving uv space radiation |
| $P_{(a)}$ | Probability of arriving at planet |

$P_{(sa)}$ Probability of surviving atmospheric entry
$P_{(r)}$ Probability of release
$P_{(g)}$ Probability of growth

(*NASA The Planetary Quarantine program; Origins and Achievements* by C. R. Phillips, NASA SP-4902)

We have serious reservations concerning this formulation and held several meetings to discuss this issue (July, August, December 1975; February 1976).

We wish to recommend the following:

## A. Inadequacy of Present "Probability Equation"

In the light of the recognition of "co-evolution," which has emerged from new advances in several fields (e.g., atmospheric chemistry, microbial ecology, microbial physiology, chemical ecology, Precambrian paleobiology, and analyses of microbial and symbiotic interactions) we conclude that the present probabilistic policy toward planetary contamination needs complete reappraisal. The assignment of numerical probabilities to the phenomena that are qualitatively unknown is inappropriate. There is limited value in the assignment of a probability between 0.0 and 1.0 of growth of a microorganism when nearly nothing is known about the identification and metabolic capabilities of the organism, the size of the initial inoculum, the presence of associated microorganisms, the details of the environment in question, and most important the detailed changes in all of the relevant environmental factors with time. Any organism leaving earth's orbit and carried on spacecraft would be subjected to changing environments during the course of its journey. Careful examination of the large number of real environmental factors that might actually determine such survival and/or growth, each of which in itself can vary greatly, would show that this superficially quantitative approach is completely inadequate. Since the probabilistic approach yields no information, it is never taken on earth in connection with growth of microorganisms; we cannot advocate it for planetary biology. The assignment of numerical values to totally unknown probabilities cannot actually dispel the ignorance inherent in this concept; it can only mislead the uninformed into believing that the values were reached by scientific and rational procedures. We believe that no such procedures are available and hence prefer to take a tempered

qualitative view. We suggest that the fundamental knowledge of the interacting nature of life on earth, the interrelations between terrestrial organisms and the continuing effects of these organisms on the atmosphere and surface of the earth ought to guide us in a formulation of a more sound scientific policy (Brock, 1973; Golubic, 1975; Cloud, 1974; Lovelock and Margulis, 1974; Margulis, 1976).

### B. The Formation of an Interdisciplinary Committee

We outline below a recommended general strategy for new policy decisions but suggest here that the detailed reassessment of present policy and formulation and implementation of new policy ought to be placed in the hands of another committee that represents at least the following fields: planetary astronomy (especially atmospheres), evolution, microbial ecology, and historical geology and biogeochemistry. Representation by those informed scientists who have designed our probabilistic present policy and/or continue to believe it valid should also be assured. Future planning should take note of a logical decision-making procedure based on the probability of considerations that were recently developed (Warner-North report).

### C. Possible General Strategy for New Policy Decisions Concerning Planetary Quarantine

The evolution of life on earth over the past 3000 million years has followed an irreversible course that has depended not only on the physical conditions at the earliest times in earth history but upon the subsequent evolution of interacting organisms. The rise of the oxygen-requiring mode of life is believed to have been only possible after the production of oxygen via microbial photosynthesis (Cloud, 1968, 1974). Numerous examples of organism mutual interdependencies and environmental interactions could be cited: coevolution of species is becoming an important principle in biology. The probability that similar events developed in detail on two planets with different starting conditions seems remote especially when it can be shown that separated populations may evolve similar structural, physiological, and behavioral adaptations (e.g., spores of bacteria and yeast; flying insects, bats and birds) but by very different routes even in organisms on the earth. That conditions throughout the flight of a spacecraft and at the planetary destination will ever be those precisely matching the survival and growth requirements of microorganisms accidently borne by such spacecraft seems to us intrinsically remote.

Furthermore, the extent to which the entire biosphere depends on its component parts has been realized lately. No known species of earth organisms can supply all of its needs for replication, growth, and distribution in the total absence of all other species. Even most strict chemoautotrophs, for example those living off only $CO_2$, salts, and water, require the continued biotic production of, for example, oxygen and hydrogen. Photoautotrophs such as blue-green algae and green plants have highly specific mineral nutrient, humidity, water activity, and incident radiation requirements. The specificity of environmental variables is determined by interaction with associated organisms (Whittaker, 1972). Heterotrophs, organisms that depend on organic nutrient sources, require that other species supply food and/or remove waste. Nearly all species require at least one type of complex organic growth factor (e.g., vitamin), and yet all vitamins are made by organisms themselves. Examples are plentiful, counterexamples not known. Based on general planetary considerations (see Section III), we believe the conditions on other planets are, and have been in the past, so unearthlike that growth of earth organisms is unlikely. Given the presently known conditions of any planets in question in the solar system with respect to parameters that may be known (for example, water abundance, incident solar radiation at liquid water level, temperature, pressure, atmospheric constituents, presence of convection and its rate and the like), COSPAR, NASA, or the SSB ought to identify those earth organisms with the highest probability of growth and survival under the known and given conditions of the planet in question. All unknown parameters may be allowed by the experimenter to vary optimally to promote growth. Any organisms that do grow under such simulated optimal conditions will be candidates for intensive experimental study in the planetary context, and specific measures for excluding given species of organisms from spacecraft might then be designed.

Because we recognize that this change of policy is at extreme variance with current practice and that this subcommittee was given explicit assignments in terms of current practice we have estimated the probability of growth for the outer planets. We find $P_g = 10^{-10}$ on Titan. See below for details.

## III. GENERAL COMMENTS ON ATMOSPHERIC MODELS FOR THE OUTER PLANETS

In order to provide a sensible procedure for discussing the possibility of growth of terrestrial organisms in the atmospheres of the outer

planets, it is necessary to consider the full range of environmental conditions that may be physically possible in those atmospheres. However, it is not necessary that a network of models, covering the full range of conditions, have been constructed; it is sufficient to concentrate attention on the possible range of models that would be most hospitable to life. When the physical environment is relatively simple, as in an atmosphere, stronger conclusions can be drawn than would be possible for a complex system, such as a land–water–atmosphere interface. When, in addition, the conditions are rather extreme, then the task of identifying conditions hospitable for life are simplified further, for the hospitable conditions themselves are likely to occupy only a small portion of the parameter range, and an extreme portion at that. It is not necessary, though it is helpful, to have made physical and chemical measurements within the environment under discussion; in their absence one must then be concerned with how well remote measurements define the environment, and what further constraints are provided by fundamental quantities such as the available energy flows. Our knowledge of the solar atmosphere depends completely on remote sensing, yet few people would argue that terrestrial organisms can replicate within it, since the environment has been rather accurately characterized and represents an inhospitable extreme.

In the case of the giant planets, elementary physical considerations applied to the measured masses and radii assure us that hydrogen and helium constitute at the least a significant fraction of the planetary mass, from at least 10 percent for Uranus and Neptune to approaching 100 percent for Jupiter and Saturn. It follows inevitably that there cannot be any "solid" interface with the atmosphere in these planets until pressures at least in the hundreds of kilobars are reached. These pressures with concomitant high temperatures are lethal to terrestrial life, and hence we need only consider atmospheric environments.

Within an atmosphere, at biologically acceptable pressure, a *sine qua non* requirement for replication is liquid water. This means that we must look for a water cloud layer in which $H_2O$ is saturated in a region −20 to 100°C. In all the models that have been constructed of outer planet atmospheres using a solar composition mixture of the lighter elements, this condition is not met, since water vapor is not saturated (e.g., water is either frozen or vaporized in these models). In models that provide water vapor saturation under liquid conditions, either the ratio $H_2O/H_2$ must be increased (by about an order

of magnitude above solar) or the temperature must be raised dramatically in the region of 1 bar pressure. Models constructed of the Jovian atmosphere to fit the remote sensing from the Pioneer 10 and 11 flybys exclude the possibility of dramatically raised temperatures. We have therefore considered only models that have increased $H_2O/H_2$ relative to solar composition. Remote sensing assures us that the temperatures at the 1 bar level in Uranus and Neptune are lower than in Jupiter. (The temperatures at the 1 bar level on Jupiter are about -100°C. The pressures in the temperature range of -20 to 100°C are likely to be more than hundreds of kilobars.)

In addition to these physical constraints, we have used a basic chemical argument about planetary atmospheric compositions. If an atmosphere must be enriched in $H_2O$, the ice presumably was added to the planet in solid form in small bodies. The enhancement of $H_2O$ does not provide an argument for a similar enhancement of $NH_3$, since $NH_3$ has a lower condensation temperature than $H_2O$. Therefore, one could only be reasonably certain that $NH_3/H_2$ would be at least the solar value upon planetary formation. But if $CH_4$, which has a still lower condensation temperature, is enhanced, then $NH_3$ must also be enhanced.* Hence ammonia is likely to be in high concentration at the water levels.

A further constraint we have used is that the atmospheres of the outer planets are convective, and hence that they are well mixed. There is good observational evidence for this for Jupiter and Saturn, since these planets emit more energy than they receive from the sun. For Uranus and Neptune, these heat flows from the deep interior have not been observationally established. However, their interiors are of a sufficiently high density to require the presence of heavier elements, and if the heavier elements and compounds including $H_2O$, $NH_3$, and $CH_4$ are in solar proportions, then there are about four earth masses of rocky materials in the core of each planet. The radioactive content of such rocky material is far more than enough to provide a heat flow sufficient to drive convection.

These physical constraints provide only a very narrow range of possibility for constructing atmospheric models hospitable for life. We do not advance the models used in our discussions as probable models; they are, in general, improbable models. But a conservative approach to the quarantine problem requires that we deliberately seek conceptual models most hospitable for life. As it happens,

*Solar value of elements (relative numbers of atoms to silicon): H, $3.18 \times 10^{10}$; C, $1.18 \times 10^{7}$; O, $2.15 \times 10^{7}$; He, $2.2 \times 10^{9}$; N, $3.7 \times 10^{6}$.

models for the Jovian atmosphere (Figure C.3.1) and the Saturnian atmosphere (Figure C.3.2) that satisfied the optimum conditions and may be extrapolated to Uranus and Neptune have been constructed; they are those of Weidenschilling and Lewis (1973) discussed in more detail on the next page.

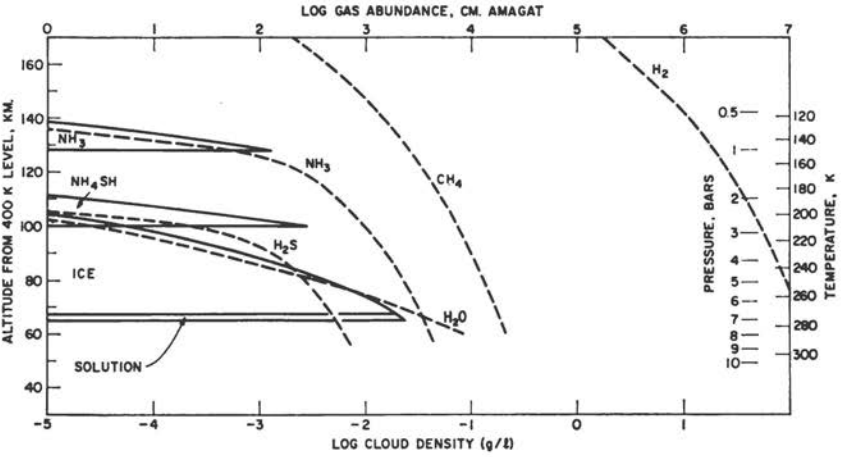


FIGURE C.3.1 Nominal atmospheric profile for solar composition of Jupiter. Solid lines show computed cloud densities; dashed lines are integrated amounts of spectroscopically active compounds in the gas phase present in a vertical column above any altitude, in cm amagat. The zero altitude is at the 400 K level. Most of the $H_2O$ forms ice clouds; aqueous $NH_3$ solution is present only marginally if supercooling is not assumed.

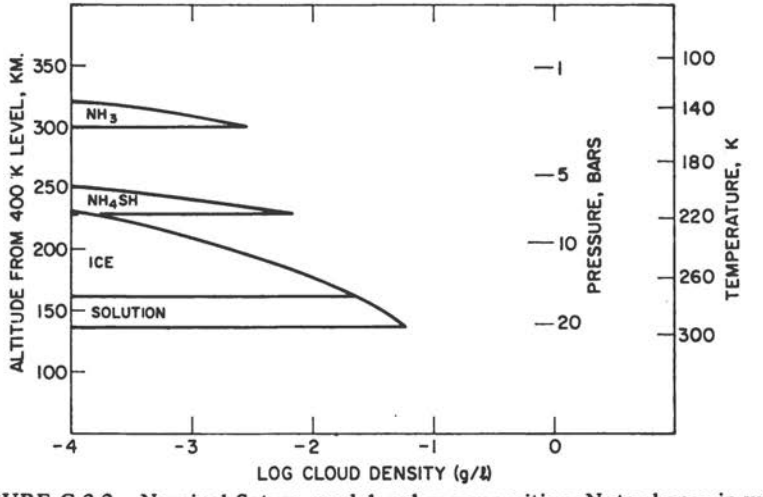


FIGURE C.3.2 Nominal Saturn model, solar composition. Note change in vertical scale.

## IV. ESTIMATIONS OF THE PROBABILITY OF GROWTH OF EARTH ORGANISMS ON THE OUTER PLANETS AND THEIR SATELLITES: URANUS, NEPTUNE, AND TITAN

### A. Probability of Growth on Uranus

The charge to the committee was to try to assess the probability of contamination of the outer planets, Uranus and Neptune, by terrestrial organisms in flyby and entry-probe planetary missions. It was concluded that the probability of growth of a microorganism on Uranus is nil, and thus the probability of contamination nil. This conclusion is based on the Weidenschilling and Lewis* (1973) model, which, of all those proposed, is most favorable for life. In this model certain elements (C, N, O, etc.) are enhanced in Uranus by a factor sufficient to form a deep, massive cloud layer of aqueous $NH_3$ solution droplets. (This model guarantees the presence of water droplets through the temperature range -20 to 100°C; other quantitative models would have the water liquid at far hotter temperatures or fail to provide liquid water throughout this temperature range.) The probability of growth was assessed with respect to a number of specific factors.

*1. Presence of Liquid Water* In the WL73 model, the base of the water cloud level is ~400 km below the tropopause at a pressure of 150 bar and a temperature of 385 K. Aqueous $NH_3$ solution clouds extend up to the 195 K level, where the pressure is 13 bar. The assumed atmospheric composition (tenfold enrichment of C, N, O, and S relative to solar abundances) is given in Table C.3.1. Figure C.3.3 shows the phase relations of the $H_2O$–$NH_3$ system over a wide range of partial pressures of $H_2O$ and $NH_3$ and of temperature. Figure C.3.4 gives the cloud structure for the nominal Uranus model of WL73. Note that the Uranus model atmosphere (line E in Figure C.3.3) shows dissolved $NH_3$ concentrations ranging from 0.3 *M* (385 K, 150 bar) to 1 *M* (335 K, 80 bar), to 7 *M* (273 K, 40 bar) to 34 *M* (195 K, 13 bar). Microbial growth has not been observed at these ammonia concentrations; evidence suggests that they would be lethal. To have a finite probability of growth a viable terrestrial organism must be either deposited in the region of liquid water stability or be transported thence by atmospheric motions. We know of no

*Abbreviated (WL73).

mechanism that might permit anaerobic organisms from the interior of a spacecraft to survive entry and be specifically deposited at the appropriate water layer. Even dropped at the top of the atmosphere by unknown mechanisms such organisms would spend perhaps weeks

TABLE C.3.1 Uranus and Neptune Atmospheric Composition Model

| Species | Volume Fraction |
|---|---|
| $H_2$ | 85.9% |
| He | 12.0% |
| $H_2O$ | 1.20% |
| $CH_4$ | 0.66% |
| $NH_3$ | 0.22% |
| $H_2S$ | 300 ppm |
| Ne | 200 ppm |
| Ar | 6.0 ppm |
| $PH_3$ | 6.0 ppm |
| HCl | 3.6 ppm |
| HF | 1.8 ppm |

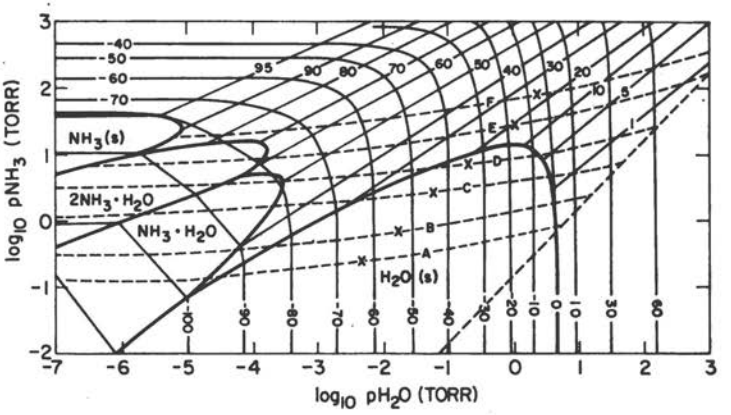


FIGURE C.3.3 A representation of the phase diagram of the $NH_3$-$H_2O$ system on the log $P_{NH_3}$-log $P_{H_2O}$ plane. The scalloped line running from far left to bottom right is the freezing line of aqueous solutions. The diagonal straight lines are solution isopleths with concentrations given in mole percent $NH_3$. The family of curves intersecting the isopleths is a set of isotherms, with temperatures in degrees C. The diagonal dashed line at the right represents the bulk $NH_3$:$H_2O$ ratio in a solar-composition atmosphere. The dashed lines labeled A through F are, respectively, cloud composition tracks for solar composition Jupiter, Saturn, Uranus, and Neptune model atmospheres and Uranus and Neptune atmospheres enriched in both $NH_3$ and $H_2O$ by a factor of ten. The x on each track marks the beginning of $NH_4SH$ formation.

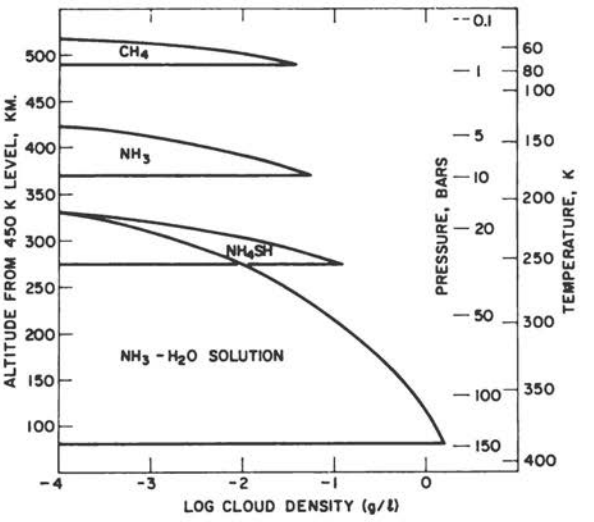


FIGURE C.3.4 Nominal Uranus model, ten times solar amounts of $H_2O$, $NH_3$, $H_2S$, and $CH_4$.

settling in the hostile atmosphere to reach the water saturated level. We do not know of spore tolerance studies of organisms subjected for weeks to high ammonia levels at varying pressures and temperatures; perhaps such studies ought to be undertaken. Assuming the microbes reached the water saturation level, the water particles to be gravitationally stable would be expected to be small, maximally $<100$ $\mu$m in diameter. No known terrestrial organism can grow in such particles, few survive more than some days in water droplets of that size suspended in a gaseous medium. The type of stability required for an indefinitely viable suspension has not been observed on the earth. (R. L. Dimmick, personal communication.)

*2. Convection* A terrestrial organism suspended in that portion of the Uranus atmosphere most favorable to growth would be rapidly convected to lower depths and higher (lethal) temperatures. The time scale for this "self-sterilization" is dependent upon the magnitude of the heat flux coming out of the deep interior of Uranus. There are several models of the interior and resulting convection of the Uranus atmosphere. Based on mixing length theory, using very conservative considerations of the convection rates Lewis (1976) concludes the mean lifetime of a parcel of atmosphere before sterilization due to convection to lethal depths is from hours to several months. We

consider the chances are low that an organism can replicate to produce enough offspring to overcome the descent of these parcels of air. Hence on any model of the outer planet's atmosphere, convection is another adverse factor for terrestrial contamination. That is, to stay viable, an organism would have to evolve a mechanism to compensate for the convection. In the probable absence of either a photosynthetic or heterotrophic source of energy for even a single cell division, physiological adaptation or evolution to convective atmospheric existence could not possibly occur before the organism is swept to depths bringing it to lethal temperatures.

*3. Energy Sources and Nutrients* The sunlight (photon flux) reaching the outer surface of the planet's atmosphere is approximately 1/400 the quantity reaching the earth. At levels in the atmosphere where water is liquid, the sunlight in the visible region of the spectrum is many orders of magnitude lower than on the earth because of scattering and absorption by atmospheric gases. The radiation field is probably in thermodynamic equilibrium with the atmosphere. Thus the quantity and quality of radiant energy at the water droplet layer would preclude photosynthesis. The hydrogen, helium, methane, ammonia water envelope could, of course, contain no free oxygen. Thus most chemoautotrophy, which in all cases studied with a single exception (Panganaban *et al.*, 1976) depends on the presence of free oxygen, is also precluded. Since the mole fraction of $CO_2$ at the water layer is expected on the WL73 model to be less than $10^{-10}$, autotrophy depending on that source of carbon can also be ruled out. Thus all modes of biological energy production would be impossible on these planets.

We considered the possibility that biologically appropriate organic compounds could be produced in sufficient concentrations by solar uv and lightning and transported to lower depths to serve as energy sources for organisms. Firstly, the probability of a qualitatively appropriate composition of organics being transported to the water cloud level in quantities neither too toxic nor too dilute itself approaches zero. All organisms found so far to contaminate spacecraft (Puleo *et al.*, 1976) require organic compounds in relatively large quantities. Many microbes require specific compounds such as vitamins in trace quantities. Lewis (1976) on the basis of several assumptions has calculated upper limits for the amounts of organic matter in the atmosphere or Uranus produced by solar uv and electric discharge. The mole fraction of total organics expected

by discharge was about $4 \times 10^{-13}$. The compounds thought most likely to form were simple hydrocarbons such as ethane and perhaps acetylene and propane: terrestrial anaerobic organisms for which such compounds provide the sole source of both energy and carbon have never been reported. None lacking the organic phosphates (ATP, GTP, etc.) are known. It seems that heterotrophic growth would not be possible either.

*4. Ammonia and Hydrogen* Virtually all models of Uranus and Neptune predict high quantities of ammonia where the temperatures are high enough to permit its presence in gaseous form. This condition of high temperature, of course, would have to be met if water were in liquid form. The ammonia concentration high in the water clouds on the extrapolated WL73 model is expected to be $>7$ *M*, which precludes the survival of all known microbes, in the growing state. At the base of the water clouds, with optimal assumptions, the ammonia concentration might be as low as 0.3 *M*. (The corresponding temperature here is 112°C.) The presently known upper limit to growth with the possible exception of the Kakalsikia-like microbe (Siegel, 1976) for ammonia-tolerant bacteria is close to 0.01 *M*, where the ionization of $NH_3$ affords the major control of pH (Deal *et al.*, 1975). Terrestrial contaminants would, of course, have to pass through and survive the higher ammonia concentrations at greater altitudes in order to arrive at this level. More information on ammonia tolerance of bacterial spores under simulated outer-planetary conditions should be obtained. It must be noted, however, that the radio brightness spectrum of Uranus and Neptune is best interpreted by using only $H_2$ (and perhaps $H_2O$) opacity. $NH_3$ has not been detected (Danielson, 1975). This probably implies low temperatures; it certainly suggests high $H_2$ concentrations. Organisms may be sensitive to $H_2$ concentrations in the absence of an oxidizing agent; for example the very primitive "S" organisms [in the *Methanobacillus omelianski* association is inhibited by its own $H_2$ production at these concentrations (Reddy *et al.*, 1972)]. The absolute limits of growth of anaerobic microorganisms in the presence of $H_2$ perhaps deserve further study.

*5. Metal Ions* K, Na, Mg, Mn, Ca, Fe, Mo, Co, and others are required in appropriate concentrations for all microbes (to maintain integrity of macromolecules and their enzymatic functioning) (Epstein, 1965; Watson, 1976). There is no conceivable mechanism to

supply all these ions in aqueous solution at the water cloud level in appropriate concentrations.

*6. Lack of a Surface* A rocky-frozen gas surface of the planet is expected at a temperature of about 3000 K, a pressure of 6 million bars, and a 8000-km depth, if it exists at all (Podolak and Cameron, 1974). Thus, it is inconceivable that optimal stable environments, where there is interaction of atmospheric gases, surface substrata, sunlight, and open bodies of water, exist.

*7. The WL73 Model and others* (Danielson *et al.*, 1973) lead to the conclusion that no known species of earth organism can survive and *grow* under Uranian conditions.

## B. Probability of Growth on Neptune

Because of its even greater distance from the sun and probably common origin, the arguments applied to Uranus can be extrapolated to Neptune (see Figure C.3.5). We see no reason to expect the probability of growth of a terrestrial organism to be anything but lower on

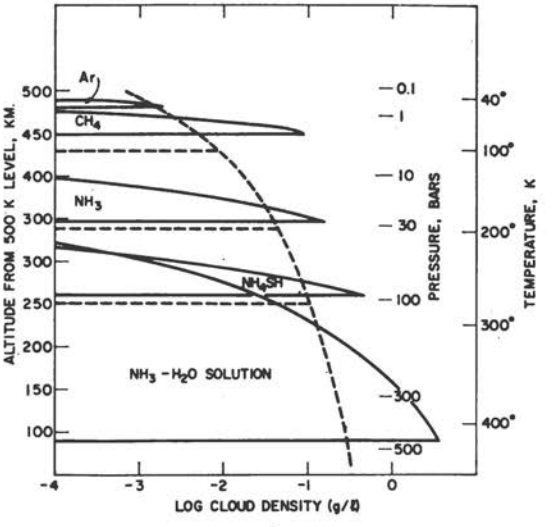


FIGURE C.3.5 Nominal Neptune model, ten times solar amounts of $H_2O$, $NH_3$, $H_2S$, $CH_4$, and Ar. Solid lines are computed cloud densities. Curved dashed line at right is maximum cloud density, if proportional to atmospheric density, and set at $10^{-3}$ g/liter at standard density. Horizontal dashed lines show schematically the lowering of cloud bases by precipitation.

Neptune than Uranus. The colder temperature and lower solar flux would increase the difficulty of growth of a viable organism delivered to the water cloud layer.

## C. Probability of Contamination of Titan

Titan, the largest satellite of Saturn, has a massive methane-containing atmosphere whose true extent is unknown. Inferences of atmospheric depth and radio occultation measurement of surface radius must await the Mariner Jupiter Saturn (MJS 1980–81) flyby. Recognizing that we are ignorant of conditions of Titan and that this satellite is of great potential interest from the point of view of prebiotic chemistry, we have, in any event, attempted to estimate the probability of contamination of the satellite by growth of a terrestrial microorganism.

Two very different models for the atmosphere of Titan are currently being considered. One of these (Danielson *et al.*, 1973) pictures extremely hostile surface conditions (90 K, direct exposure to solar uv of $\lambda > 1650$ Å). If the Danielson *et al.* model is valid, the probability of growth is highest at the upper limit of the greenhouse effect, where the temperature might be as high as 130 K. Danielson claims, "The present radio brightness temperatures limit the surface temperature to 100 ± 36 K" (letter, February 9, 1976). The lowest temperature for any biological growth process recorded is 253 K. The lowest temperature at which liquid water can be detected in biological systems is about 200 K (Mazur, 1970). The lowest temperature for liquid water under any circumstances is about 130 K (McMillan and Lus, 1965; Yannas, 1968). Clearly, water-based life as we know it on earth is precluded by Danielson's model.

The other model that shall concern us here permits far warmer surface conditions and uv shielding, possibly even allowing a surface ocean of aqueous $NH_3$ solution (Hunten, 1972; Lewis and Prinn, 1973; Pollack, 1973). In the latter model, a large greenhouse effect might raise the surface temperature from the gray-body equilibrium value of ~80 K to 160 K or higher. If the surface of Titan lies far enough below the spectroscopically observed level, aqueous $NH_3$ solution may be present on the surface ($T \geqslant 173$ K; $P = 1.0$ to 20 bar): a surface $CH_4$ partial pressure less than 0.8 to 1.0 bar would be incompatible with temperatures above the $NH_3$–$H_2O$ binary eutectic. (See Figure C.3.6.) If such surface melting occurs, then the liquid composition will be very close to that of the eutectic melt, 20

*M*. If the surface temperature is well above the eutectic temperature, then the $NH_3$ concentration could become as low as that given by the cosmic abundance ratio of N to O, about 8 *M*. This ammonia concentration is lethal in all cases studied.

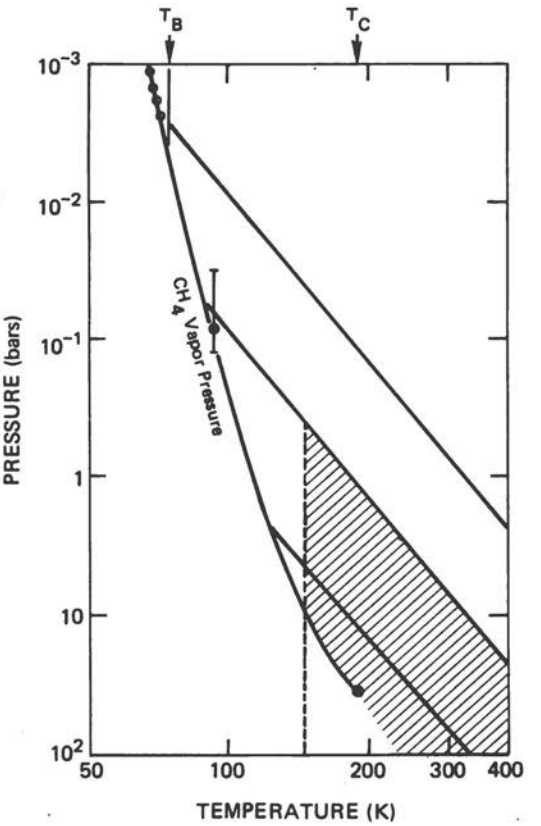


FIGURE C.3.6 Pure-methane atmospheric models for Titan. The heavy line is the vapor pressure curve of methane. $T_B$ is the Gold-Humphreys boundary temperature (74 K), and the $T_C$ is the critical temperature of methane. The triple point of methane is indicated at 91 K and 90 mbar, in good accord with the effective temperature and $CH_4$ pressure for a pure-methane atmospheric model. The three diagonal lines are dry adiabats for pure $CH_4$. Note that the slope of the vapor pressure curve (a fully saturated adiabat) equals that of a dry adiabat near the critical point. The cross-hatched region contains the allowable surface conditions ($T_S \geqslant 145$ K) for pure-$CH_4$ atmospheres. The visible level in Titan's atmosphere is presumably defined by the triple point of methane, above which level a bright solid $CH_4$ particulate haze may be present but below which only strongly forward-scattering liquid droplets would be stable.

We have also considered a Titan model designed to have the lowest reasonably defensible ammonia concentration. Our lower limit to the ammonia abundance on Titan has been estimated by the following procedure. First, it is assumed that Titan accreted largely from material totally devoid of methane and ammonia. Second, the methane and ammonia actually present in Titan are assumed to result from "contamination" of Titan by a small amount of icy material containing solid hydrates of methane and ammonia. Third, the minimum amount of methane necessary to produce biologically interesting surface temperatures ($\geqslant$273 K) is calculated for a dry adiabatic pure-$CH_4$ atmosphere. The minimum $NH_3$ content then follows. Fourth, we assume the $NH_3$ is uniformly distributed throughout the "icy" part of Titan (not enhanced at the surface because of its volatility).

The minimum $CH_4$ pressure is found to be 7 bar, giving a total mass of $5 \times 10^{22}$ g of methane. In an ice condensate formed at low enough temperature to retain $NH_3$ and $CH_4$ as solid hydrates, the $NH_3/CH_4$ mass ratio is ~2. The bulk density of Titan (1.5 g/cm$^3$) requires that low-temperature condensates occupy ~70 percent of the volume of Titan; thus, the mass of ammonia per unit mass solution is found to be $1.4 \times 10^{-4}$, equivalent to a solution concentration of 0.008 *M*. This degree of dilution is that obtainable if *all* ice is melted in the interior of Titan. Thus, even after a series of conservative assumptions, the lower limit on the $NH_3$ concentration is found to be essentially identical with the *upper* limit on $NH_3$ concentrations tolerated by the most ammonia-tolerant terrestrial microorganisms so far studied (Deal *et al.*, 1975).

A more reasonable interpretation of the low density of Titan and the presence of methane would be that the formation temperature of Titan was low enough for total retention of $NH_3$ and $H_2O$ in solar proportions. As we have seen, this would require an $NH_3$ concentration of ~8 *M*.

Molecular oxygen would in any case be absent, and lack of mineral nutrients would provide a further constraint on the probability of growth. Bacteria on the earth found in environments where methane is present include methane oxidizers (which generally *require* molecular oxygen and hence would be precluded) and methanogenic bacteria. Panganaban *et al.* (1976) have recently isolated an anaerobic strain that grows on methane using sulfate to oxidize it as a source of energy. The ammonia tolerance of the organism is unknown and should be investigated. However, methane producers require $CO_2$ and $H_2$ and are usually found in environments rich in organic matter

such as lake bottoms and sewage. Methane-producing anaerobic bacteria comprise a small fraction (perhaps $<1/10^4$) of the total microbial diversity; given their ecology, their initial presence on a spacecraft is unlikely. Growth of organisms at 1 to 20 bars of methane at 8 *M* ammonia concentrations is not known, but perhaps Titan simulation growth experiments ought to be encouraged. It should be noted that many attempts to grow methane-producing bacteria on the earth under what are planned to be optimal conditions have often failed. Unlike the situation on Titan, in these experiments certainly water and temperature conditions are not severely limiting.

Recognizing both the interest in possible organic chemical reactions on Titan and that the probability of growth of a terrestrial microbe is likely to be vanishingly small, we suggest that a $P_{(g)}$ of less than $10^{-10}$ is adequate until revisions are required on the basis of MJS results. (We estimate the probabilities as follows: anaerobic, $10^{-1}$; high-methane low-temperature tolerance as well as the presence of growth factors necessary for methane bacteria, $10^{-5}$; tolerance to 8 *M* ammonia, $<10^{-4}$.) If observations show that ammonia has been converted to $N_2$ on Titan, the chance of growth of microbes would have to be re-evaluated. That is, although at present $10^{-10}$ is probably a conservative estimate of the probability of growth, we want to leave it high so that it will be adequately re-evaluated as the models for the enigmatic Titan become refined and more data become available.

## V. GENERAL LIMITS TO GROWTH OF EARTH ORGANISMS

We include here some information, compiled primarily by Peter Mazur (Spring, 1975), adapted from a letter written by Mazur to A. G. W. Cameron.

In general, if the following conditions are not met, the probability of growth of a terrestrial microorganism on another planet would approach zero. These conditions which set limits to growth are summarized in Table C.3.2, but some of the points need amplification. For example, in Table C.3.2 the column headed "survival" becomes important if terrestrial contaminants find themselves in oases favorable for growth only a portion of the time. Only vegetative cells are capable of growing. Data are given for survival of spores because they are more resistant, and one could conjure up the (unlikely) scenario of oases becoming hostile slowly enough and under the proper conditions to permit the vegetative cells to convert to spores, which if and

TABLE C.3.2 Approximate Limits for Growth and Survival of Terrestrial Microorganisms

| Factor | Limits for Growth | Limits for Survival (1-h Exposure) | | References |
|---|---|---|---|---|
| | | Vegetative Cells | Spores | |
| Temperature | >−15°C<br><85°C | <85°C<br><4 K[b] | <160°C<br><4 K[b] | Brock, 1966; Emerson, 1968; Ernst, 1968; Farrell and Rose, 1967; Fogg, 1969; Pflug and Schmidt, 1968; Porter, 1945 |
| Water activity | >0.9[a] | 0 to 1 | 0 to 1 | Beers, 1957; Cochrane, 1958; Fogg, 1969; Horowitz *et al.*, 1972; Charling and Horowitz, 1974; Ingram, 1957; Porter, 1945; Scott, 1957; Webb, 1965 |
| Pressure | 600 bar | 3000 bar | 20,000 bar | Johnson, 1957; Zobell, 1970 |
| pH | 11.5 | 12 | Not known | Deal *et al.*, 1975; Porter, 1945; Souza *et al.*, 1975; Thimann, 1963 |
| Uv radiation (2600 Å) | – | 0.1 J/cm$^2$ | 0.1 J/cm$^2$ | Donnellan and Stafford, 1968; Schechemeister, 1968 |
| Ionizing radiation | – | 2–4 Mrad | 2–4 Mrad | Goldblith, 1953; Silverman and Sinskey, 1968 |
| Nutrients | See text | | | |

[a] >0.6 for certain fungi[9]; >0.8 for halophiles.
[b] No lower limit has been reported.

when they again find themselves in oasis conditions, germinate to reform vegetative cells.* Requirements for survival would, of course, become critical if oases exist only in the atmosphere, and especially if the atmosphere is convective.

Now to comment on some of the individual physical and chemical factors.

*Induction of spore formation actually occurs in only a fraction of the total microbiota, only those genetically capable, and is brought about by specific inductive mechanisms that would tend to be unlikely on other planets.

*1. Temperature* The difference between upper limits for growth and the upper limits for survival is relatively small. The stated upper limit for growth may not be the absolute limit (growth at 104°C, 1000 bar has been reported) (for review see Brock 1966, 1973), but it probably is close to a realistic limit for the organisms contaminating spacecraft. At the low end, organisms can survive to 1 K, but each cycle of exposure to freezing would kill a fraction, the value of which depends on the organism. In their extensive review of minimum growth temperatures, Michener and Elliott (1964) report that microbial growth below −10°C is rare. Three reports of growth from −17°C to −34°C have not been confirmed. The best evidence that growth does not generally occur in these temperatures is that billions of cartons of frozen food are stored at −18°C without spoilage. Siegel *et al.* (1968) reported penicillium spores germinated and incorporated nucleotides and amino acids in the liquid ammonia-glycerol, but as Deal *et al.* (1975) point out, the significance of "germination" is difficult to determine. No claim was made that the germinated hyphae survived. Morita (1975) has extensively reviewed the literature on low-temperature bacteria. The polar and ocean regions of the earth constitute about 85 percent of the surface, and more than 90 percent (by volume) of the ocean is 5°C. Although many cold-growing (psychrotrophic) and cold-loving (psychrophilic) strains of bacteria have been isolated, the lower end of the thermal spectrum of these two classes of organisms was 4 to 5°C for the former and −4 to −5°C for the latter. There is no doubt that the organisms tested are sensitive to temperatures outside those to which they are optimally adapted. The psychrophiles tend to be sensitive to even moderate temperatures; they leak both small and macromolecules into the medium. They are also very sensitive to other environmental variables; e.g., nutrient concentrations, salinity, and hydrostatic pressures.

*2. Water Activity** The reason for giving two limiting values for growth are the following. About the only bacteria that grow at water activities <0.9 are the halophiles and some fungi. The former are highly specialized and *require* high concentrations of NaCl to grow. Growth generally requires $O_2$ (Raymond and Sistrom, 1969). The growth of most fungi requires oxygen, and $O_2$ is not believed to be

*$a_w$ is water activity. $a_w = P/P_0$, where $P$ is the vapor pressure of water in a solution and $P_0$ is the vapor pressure of pure water.

present on the outer planets. Only one anaerobic halophile is known, and this organism requires complex growth factors and visible light (Horowitz, personal communication). Even if the proper $a_w$ is available, growth seems to require the presence of liquid water–high humidity vapor is not sufficient. There have been a few unconfirmed claims that fungi may be an exception, but as indicated, nearly all fungi can be excluded on the basis of the absence of oxygen.

*3. Pressure* The limits for growth and survival are rather broad. Actually Zobell has reported growth at 1400 atm in organisms isolated from ocean depths, but he comments that organisms will not grow when pressures exceed the ambient by ~500 atm. Microorganisms from ocean depths would seem unlikely contaminants on spacecraft.

*4. pH and Ammonia* The presence or absence of $NH_3$ in the oasis is critical with respect to pH. The pH's indicated are absolute cutoffs, but published data indicate that very few organisms can grow above pH 9 or 10, although growth for at least one to pH 11.4 has been reported (Deal *et al.*, 1975). For example, Mono Lake (California) has a pH of 10.5 yet supports a rich microflora. Ammonia itself, however, is generally toxic, the upper limit found by Deal *et al.* (1975) was $< 0.01$ *M*. Further research on ammonia and pH tolerance under simulated outer planetary conditions is warranted.

5. *Uv Radiation* The figure given ($3 \times 10^{-5}$ J/sec/cm$^2$) is the uv flux on hospital operating tables that essentially eliminates infection of patients. It seems like a reasonable upper-limit value. This flux will kill the most resistant of terrestrial microorganisms in from 0.2 to 2 h. However, the problem of uv killing is complex both physically (shielding by uv-reflecting or -absorbing materials) and biologically (biological repair).

*6. Ionizing Radiation* Especially important here would be the killing of organisms on the spacecraft by solar wind, cosmic rays, and the planet's radiation belt during the 4 to $\geqslant$9 year traverse to the planet.

*7. Nutrition* Required elements include C, H, O, N, P, S, Na, K, Mg, Mn, Ca, and Fe. These have to be present in a form to be utilizable by organisms (e.g., N. Horowitz argued that phosphine has

not been shown to be utilized by anaerobes (but see Foster, 1976). This is an area where further experimentation is necessary. Another important point is that nutrients be present in sufficient but not toxic concentrations. Even in terrestrial oceans and lakes nutrient elements are limiting and populations rarely exceed $10^6$ organisms/ml.

*8. Convection* On the outer planets, rapid transfer of organisms to lethal zones would preclude growth. But even if such convection is slow or absent, formidable barriers to growth remain. In spite of the fact that much of the earth is an oasis, Brock states, "It is likely that little or no growth of microorganisms occurs in the air itself, the air functioning only as an agent of dispersal."

*9.* $a_w$ As mentioned, in order to grow, microorganisms must apparently be in liquid water of $a_w \geq 0.9$. To be gravitationally stable, the liquid water in the atmosphere would have to be in small drops. But small drops have a very ephemeral existence. Webb (1965) reports that the 10-$\mu$m drops of water at 80 percent relative humidity evaporate in fractions of a second. Individual microorganisms exposed to low relative humidities will equilibrate in milliseconds, and each cycle of drying will kill a considerable fraction of vegetative cells. A scenario for growth in the atmosphere would seem to require relative humidities $\geq$ 95 percent most of the time for long-lived drops, or, even more unlikely, a high probability that an organism that has its surrounding droplet evaporated will find residence in another droplet in a fraction of a second.

*10. Simultaneity of Required Factors* Table C.3.2 lists upper limits for each factor individually, but growth requires that appropriate values for all the factors be present simultaneously. The limits for growth become considerably narrower in the latter case. For example, the SSB *ad hoc* Review Group in 1970 defined the minimum conditions for an oasis capable of supporting the growth of terrestrial organisms to be:

A. $a_w \geq 0.95$.

B. *Temperature*: $\geq 0°C$ for 0.5 h per day (an upper temperature limit was not given since this was for Mars).

C. *pH*: 5 to 8.

D. *Nutrients*: small amounts of water-soluble appropriate compounds containing N, S, P, and C (and/or light and the proper gases).

And it stated, "All of the above occurring simultaneously or nearly so."

*11. Relations with Other Organisms* In order to initiate growth to high density, very often certain minimal inoculum sizes (e.g., initial number of individuals) are required. Nearly all (perhaps all) earth microorganisms depend on members of other species for removal of waste, production of food sources, dissemination, delivery of nutrients, and so forth. The probability of the initiation of growth by minute populations tends to be low even on the earth.

### Summary of Conclusions Concerning Growth of Earth Organisms on Outer Planets

The outer planets and their atmospheres constitute an environment hostile to terrestrial microorganisms. Among the reasons for this are the following:

(a) Absence of liquid water on a planetary surface; prohibitive temperatures and pressures at the planetary surface if it exists at all.
(b) Atmospheric convection that would tend to transfer organisms to lethal zones.
(c) Paucity of energy sources for photo-autotrophs.
(d) Lack of organic substrates for heterotrophic growth.
(e) Lack of oxygen (or other oxidants) for chemoautotrophic growth.
(f) Lack of ions in appropriate concentrations for growth.

These considerations, taken in combination and summation, exclude the possibility for growth of any known terrestrial organisms surviving transfer to the outer planets by spacecraft.

## VI. SUMMARY OF SUGGESTED EXPERIMENTS

In preparing this report on the chances of growth and survival of organisms on outer planets we have noted some glaring deficiencies in fundamental knowledge. More information on the following would be of interest to both planetary and earth-based biology. Thus we recommend these further terrestrial experiments:

1. Determination of the upper limits of growth and spore survival to various hydrogen concentrations and pressures.

2. Search for microorganisms that can utilize phosphine as a sole source of phosphorous.

3. A survey of the sensitivity of microorganisms to phosphine.

4. A search for anaerobic halophiles and a determination of their tolerance to low water activities should be undertaken.

5. Determination of the upper limits of growth and spore survival to various concentrations and pressures of other gases likely to prevail in the envelopes of the outer planets, e.g., hydrogen sulfide, methane, and ammonia.

6. The tolerance of the recently isolated anaerobic methane oxidizing bacteria to simulated Titan conditions ought to be investigated. We also note that the information available on the microelement nutrient requirements of microorganisms at the lower temperature limits for growth is limited. The acquisition of more such information might be of general interest.

7. Further investigation on the growth of microorganisms in water droplets suspended in atmospheres ought to be undertaken.

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## ACKNOWLEDGMENTS

We acknowledge with gratitude critical input by Ronald Prinn and Peter Stone, Department of Earth and Planetary Sciences (MIT); we further acknowledge the review and criticism of our colleagues Peter Mazur, Division of Biology, Oak Ridge National Laboratory; I. R. Kaplan, Department of Geology, UCLA; Thomas Jukes, Space Science Laboratory, University of California; E. S. Barghoorn, Harvard University; C. D. Cox, University of Massachusetts; Norman Horowitz, Department of Biology, California Institute of Technology; G. J. Wasserburg, Department of Geology, California Institute of Technology.

Critical helpful comments on early drafts were received from J. Lederberg, E. Levinthal, C. Sagan, R. L. Dimmick, R. S. Young, A. Ingersoll, C. Leovy, P. H. Deal, S. Rittenberg, R. Hansen, T. Owen, and C. S. Cox.

### Attachment C.4

January 22, 1976

Dr. Peter Mazur
Biology Division
Oak Ridge National Laboratory
Union Carbide Corporation
P.O. Box Y
Oak Ridge, TN 37830

Dear Peter:

In response to your letter dated January 2, 1976, I am sending you herewith some comments concerning NASA experiences with various planetary missions and the impact of planetary quarantine requirements on costs and reliability. I hope that this information will provide a valuable background during your formulation of policy recommendations on outer planet quarantine.

For planetary quarantine purposes, planetary missions can be classed as flybys, orbiters and landers. The early Mariners intended for only brief planetary encounter are examples of flybys. The more recent Mariner Mars '71, intended for planetary encounter, orbit, and extended operations is an example of an orbiter. The current Viking is an example of a lander mission.

The various options to reduce bioloads on spacecraft that have been used to date are as follows:

1. (used for flybys) operations conducted in an environmentally defined clean room with biological monitoring and assay performed. Thus the bioload at time of launch can be defined.

2. (used with orbiters) in order to insure that a specified bioload is not exceeded, measures under option one are expanded to include spacecraft cleaning and microbial load reduction.

3. (used with landers) when the specified bioload is very low, then measures from options one and two are expanded to include terminal sterilization.

The term sterilization as used in the NASA planetary quarantine program is defined as a microbial load reduction procedure, whereby a known microbial load is reduced to a specified level, i.e., subjecting a spacecraft to a dry heat environment for a given time. The matter of whether piece parts and components must receive separate treatment is related to how the third option is to be achieved. Although many methods other than dry heat sterilization have been examined, no alternate has yet been found that is both effective in reducing microbial load and acceptable for general spacecraft application.

Experiences with the Mariners (flyby and orbiter) are now sufficiently well documented to permit conclusions concerning the impact of planetary quarantine on both costs and reliability (see enclosure 1). NASA planetary quarantine *research* costs during the period FY'67 through FY'75 amounted to about 1.2% of all costs for planetary flight projects.* Planetary quarantine *project* costs for the Mariners during the same period amounted to about 0.2% of costs for the Mariner projects. There was no case of component part or spacecraft failure traceable to quarantine requirements. It is generally believed that the required cleanroom procedures and environmental control lead to an increase in reliability. In order to achieve spacecraft reliability, most of the contamination control procedures used for the Mariners would, in fact, be continued even though quarantine requirements are not imposed.

Although the Viking mission (orbiter and lander) is not yet complete, experience gained through the development, test and launch of the two sets of flight hardware permits an early estimate of added costs due to planetary quarantine requirements. The requirement for the completed landers to undergo a dry heat sterilization treatment resulted in increase in costs for selection, test, and processing of materials and parts, as well as, design fabrication and check out of sterilization facilities. In addition special poststerilization requirements, such as the bio-shields for the landers increased design and fabrication costs. It is estimated

*Launch vehicle costs, as well as tracking and data acquisition costs, were not included in project costs. If they had been included, the percentages for planetary quarantine would have been lower.

that such costs amounted to about 5% of the total for Viking. It is felt that since the heat requirements were part of the initial design requirements and the design and test programs implemented, there was no impact on reliability of Viking hardware performance. The heat sterilization was also required to insure that the lander science will not show false indications due to possible earth contamination. In fact, it was this requirement that determined the duration of the Viking terminal sterilization cycle.

Considering the effect of various values for the probability of growth ($P_g$) on the requirements to sterilize, it should be noted that a very low value ($10^{-6}$) was specified for Mars but dry heat sterilization was still required for a probability of contamination ($Pc$) of $10^{-4}$ per launch. Some lower value for $P_g$, perhaps $10^{-8}$ or $10^{-9}$, would no doubt permit a lander mission without the requirement for heat sterilization. It is by no means clear at what point the value of $Pg$ would be sufficiently low as to permit a lander or probe mission without sterilization. One approach would be to use a general equation $Pc = N_O Pg^*$ as a rough order of magnitude estimate. I expect the matter of whether or not a planet or body is considered to be of biological interest will be of prime concern when it is necessary to specify quarantine options such as terminal sterilization and also assist in determining $Pc$ for the planet, as well as suballocations to the various missions. Mr. Leo Daspit has recent experience in the use of quarantine models and suballocations for the Viking mission. I think it would be advantageous to have him review his experiences with the panel. If you agree, I will take steps to invite Mr. Daspit to the meeting scheduled for 11 March 1976.

Sincerely,

Richard S. Young
Chief, Planetary Biology and Quarantine
Office of Space Science
National Aeronautics and Space Administration
Washington, D.C. 20546

Encl: JPL Planetary Quarantine Cost Study 1975

*$Pc$ = probability of contamination suballocation to lander or probe. (In the case of Viking, this suballocation was $\sim 2 \times 10^{-5}$.)
$N_O$ = number of surface organisms on lander or probe.
$Pg$ = probability of growth.

# Appendix D

February 4, 1977

Dr. Alastair G. W. Cameron
Chairman
Space Science Board
Center for Astrophysics
Cambridge, MA 02138

Dear Al:

The Space Science Board, and especially the Mazur Committee, have done a valuable service to reassessing the growth probability for terrestrial organisms on Uranus and Neptune. There are still a couple of loose ends that could, and should, be tied off with a modest further effort. If this is not done, I see possible impacts on the Viking Orbiter extended mission and on the design, reliability, and cost of the Jupiter Orbiter with Probe (JOP). While these impacts cannot readily be documented at present, it is my opinion that they may be significant. The two areas that call for attention are:

(1) *Mars*. We have information, both empirical and theoretical, that the surface environment of Mars is extremely hostile. The presence of powerful oxidants in the soil is indicated at two widely separated locations. Organic matter is absent or extremely scarce. Both can be understood in terms of the action of solar uv on water molecules in the atmosphere (Hunten) and adsorbed on surfaces (Huguenin). These models strongly imply that the two Viking sites are typical of the whole planet. The probability of growth for terrestrial micro-organisms in such an environment should be set with the new facts and hypotheses in mind.

(2) *Jupiter and Saturn*. The arguments generated for Uranus and Neptune apply without essential change to Jupiter and Saturn. This application has not been explicitly made because it was not specifically requested. I now make the request.

If you and the Board approve, could you pass these requests to Peter Mazur for the attention of the Committee on Planetary Biology and Chemical Evolution.

Sincerely,

Noel W. Hinners
Associate Administrator
for Space Science
National Aeronautics and Space Administration
Washington, D.C. 20546

# Appendix E

February 17, 1977

Dr. Noel W. Hinners
Associate Administrator
for Space Science
Washington, D.C. 20546

Dear Noel:

This is to acknowledge receipt of your letter of February 4, requesting the Space Science Board via its Committee on Planetary Biology and Chemical Evolution to make possible revisions in quarantine statements for Mars and also for Jupiter and Saturn.

Our present plans are that the committee would produce a revised quarantine statement for Mars which the Space Science Board could adopt for its annual report in May. It should be a fairly simple matter to add Jupiter and Saturn to this charge to the committee. The committee will therefore be considering these questions in the near future and I expect it to make recommendations to the Board on these issues.

Sincerely yours,

A. G. W. Cameron
Chairman, Space Science Board

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