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THE NATIONAL ACADEMIES Advisers to the Nation on Science, Engineering, and Medicine

CONFERENCE

ON </u>

HAZARD OF PLANETARY CONTAMINATION

DUE TO

MICROBIOLOGICAL CONTAMINATION IN THE INTERIOR OF SPACECRAFT COMPONENTS

15 February 1965

Space Science Board National Academy of Sciences 2101 Constitution Avenue, N. W. Washington, D. C.

ATTENDEES:

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The sterilization of exterior surfaces of spacecraft components may be inadequate to protect the moon and planets from contamination by terrestrial microorganisms. It is possible that viable contaminants lodged within the solid materials which make up the spacecraft may be released upon or following impact. Assessment of the likelihood of contamination from this source and the study of means to prevent it were the subjects of a Conference held on July 28, 1964 by the Life Sciences Committee of the Space Science Board, National Academy of Sciences. Sixteen specialists from scientific institutions, government and universities participated.

The Conference was called following a request for advice on this matter from the National Aeronautics and Space Administration to the Space Science Board.

The Conference was asked to give general advice on the following questions:

- In what ways could microbiological contaminants in the 1. interior of spacecraft components constitute a special hazard of planetary contamination?
- In the event of a hard landing which might result in 2.1 some fragmentation of components, would internal contaminants constitute a hazard comparable to that of surface contaminants?
- 3. What means should be used to sterilize the interiors of spacecraft components, and what standards of sterility prevail?

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4. What research should be carried out to enable NASA to deal effectively with the problem of internal contaminants?

It seemed appropriate to begin discussion with a review of the rationale of spacecraft sterilization to determine whether in fact participants were agreed on the necessity of the current sterilization policy.

$1.$ The Rationale for Spacecraft Sterilization

The complex and costly process of spacecraft sterilization has been undertaken as a national policy for several compelling reasons If life does exist beyond the earth, and there is substantial evidence in favor of the hypothesis, an unsterilized or inadequately sterilized space probe could introduce terrestrial microbes, possibly causing its profound alteration or even destruction. The loss would be irreparable. Terrestrial organisms, should they be able to proliferate on the virgin surface of another planet, could lead to major alterations in the abiotic portion of the environment as we know has been the case on the earth. However, the principal reason for avoiding the introduction of terrestrial. organisms is to prevent their possible interference with life detection devices.

Of the celestial bodies immediately accessible to man, Mars is the only one where the probabilities strongly favor the existence of life. It is particularly important, therefore, that a

dedicated effort be made to retain the integrity of that life. and that, as the Space Science Board has stated, Mars should become a biological preserve. Although it is thought unlikely that the rigorous Martian environment would be conducive to the growth of many terrestrial organisms, experiments have indicated that a number of potential contaminants could survive.

The detection of extraterrestrial life has been considered sufficiently important to warrant the development and incorporation into Martian probes of extensive life detection equipment. If these experiments, or any of the payload, are not sterile, they could introduce terrestrial microorganisms into their own sample collection devices. It is also conceivable, owing to the prevalence of strong Martian winds, that a second probe landing at a site remote from a first probe could pick up terrestrial contaminants which the first had introduced. Since no way is known to distinguish in advance between terrestrial organisms and the sought for but unknown Martian life forms, any positive results from the experiments would, if any terrestrial contamination were possible, have ambiguous signif-It would indeed be ironical if the lack of sterilization icance. of the probe should frustrate the very purposes for which the probe was designed.

The Conference thus agreed that the need for spacecraft sterilization is absolute in the sense that the risk of inadvertently contaminating Mars by a non-sterile probe should be reduced to a

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negligibly low probability. While it is not possible to render a probe completely sterile, the risk can reasonably be reduced to the 1 x 10^{-4} level recommended by the international Committee on Space Research at its meeting in Florence. Italy in May 1964. The Conference fully endorsed the COSPAR position: **'COSPAR** accepts, as tentatively recommended interim objectives, a sterilization level such that the probability of a single viable organism aboard any spacecraft intended for planetary landing or atmospheric penetration would be less than 1 x 10^{-4} , and a probability limit for accidental planetary impact by unsterilized fly-by or orbiting spacecraft of 3 x 10^{-5} or less:"

2. Microorganisms in Solids

Present sterilization techniques may be inadequate to achieve the desired level of sterility for they do not allow for contaminants within the materials. When surface sterilized solids metals, plastics, circuit components, and so forth - are crushed, ground up, and placed in a culture medium, microorganisms released thereby grow and reproduce. The number of viable microorganisms in a given solid varies very greatly, depending on the organic constitutents of the solid, its toxicity, and the conditions under which it is manufactured. Experiments to date have proved unsuccessful in determining the quantity of organisms in a given solid component. The distribution of the organisms within the solid also is open to question; it can only be said that it al-

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most certainly is not uniform. Owing to the extreme difficulty of working up experimental procedures, no satisfactory method has yet been devised to determine the number and distribution of internal contaminants, nor have the governing principles been discovered. Until they are, both the assessment of the probabilities of contamination from solids and satisfactory methods of their sterilization will be largely dependent on educated guesses. It will be necessary, in order to ensure that adequate precautions are taken. consistently to assume the "worst case" - greater precautions than may in fact be warranted. The Conference believed, therefore, that the solution to this problem is worthy of a far more ambitious research effort than is now underway.

3. Likelihood of Release of Encapsulated Contaminants

The existence of viable microorganisms in solids could even so be ignored if there were assurance that the microorganisms would not be released. There are three ways in which the release might occur: 1) by the accidental fracture of spacecraft components as a result of hard landing, 2) by means of weathering or other kinds of erosion which would gradually expose the contaminants to the planetary environment, or 3) by "diffusion" or particle migration of organisms to the surface of the encapsulating components. Present information is inadequate to permit any estimate of the probabilities of release due to erosion or diffusion. Moreover, it has not been demonstrated that current methodology is adequate

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to obtain such information. These deficiencies are most serious, in the Conference's view, and render it virtually impossible to predict the degree of hazard attributable to internal contaminants, or, alternatively, to design components capable of withstanding erosion and diffusion.

In regard to the first manner of release, the likelihood of fracture upon impact depends on impact velocity, impact deceleration according to the character of the planetary surface, and, in particular, on the mechanical design of the impacting structure. Impact velocities in Mars missions will range from 10 to 500 feet per second, depending on the density of the atmosphere and the use of parachute or retro-rocket braking. It is possible to design spacecraft components which will remain intact under impacts within this velocity range, and, in special cases, even under impact decelerations of 6,000 to 10,000 g. Thus, by proper attention to mechanical design, all encapsulated microorganisms can be kept within the spacecraft components after a hard landing.

Other factors, however, bear on the problem. Firstly, the reliability of the high-impact-resistant spacecraft cannot be guaranteed to the required order of 1 x 10^{-4} , particularly in view of lack of information on critical surface conditions on which the impacts will occur. Secondly, a design which is impactresistant and capable of preventing the initial release of internal contaminants is by no means proof against erosion or diffusion.

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Thirdly, the mechanical protection against fracture postulated above does not yet obtain, and will depend on designs which are beyond the present state of the art. It should be noted that the engineering studies on fracture to date have been directed toward determining the functional integrity of components following impact rather than on the release of contaminants. Consequently, there is much still to be learned before confidence can be placed in such designs as means of reducing the likelihood of contamination.

Finally, high-impact-resistant design requirements would inevitably impose restrictions on the design of experiments landed by the spacecraft. The scientific potential of the mission would be reduced to a degree difficult to predict. In the absence of adequate information on the degree of restriction, it seems best to re-emphasize that the basic program objective is to maximize, within the limits set by the non-contamination policy, both mission success and scientific capacity of the experiments. At present it appears that this objective can more easily be met by rigorous sterilization and by retention of flexibility in the design of experiments.

4. Possible Sterilizing Effect of the Martian Diurnal Freeze-Thaw

When vegetative cells produced by spore-forming organisms, in a liquid medium, are exposed to alternate freezing and thawing, the rate of kill can easily be made to exceed the reproductive rate so that the entire population can be destroyed. Since the Martian environment includes a diurnal freeze-thaw cycle, more sensitive

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vegetative cells are not apt to survive for many days. Thus it may be argued that temperature-time requirements for sterilization can be lowered for Martian landers to a level where only the more easily killed non-spore-forming organisms are eliminated. The danger of contamination of the planet would be minimal, since it could occur only during the few days of the viability - in a weakened state of the vegetative cells; the spores, even though capable of survival for long periods of time in a dormant state since they are relatively resistant to freeze-thaw and heat, would be harmless so long as they remain dormant.

Presently available data are insufficient, the Conference believed, to substantiate the assumption on which the above argument is based. Even among vegetative cells arising from germinating spores, the lability of microorganisms to freezing and thawing varies widely. Relatively few species have been examined under well simulated Martian conditions of thermal cycling. It is important, the Conference believed, to continue the work on this problem which is now underway at NASA Ames Research Center and elsewhere. Until definitive data have been amassed to prove that the Martian environment can be depended upon to eliminate all vegetative cells, the Conference cannot regard that possibility with optimism, nor can it recommend on that basis that sterilization standards be lowered.

Dependence on the Martian environment to avoid contamination

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would introduce another, quite different, complication. If it destroyed all vegetative cells, the dormant spores could not contaminate the planet. However, the dormant spores would be present, and might be gathered into the life detection samples. Even if the environment destroyed all of the terrestrial organisms, including the spores, the operation of the life detection experiments would have to be delayed for some time until they had all been destroyed. It would be difficult, to say the least, to establish the safe minimal period with confidence. Either alternative thus appears unacceptable.

$5.$ Sterilization Standards for Internal Contaminants

Since it cannot be assumed, therefore, that some internal contaminants will not be released into the planetary environment, prevention of contamination must come from the sterilization of the solids. It further follows that the accepted standards for spacecraft sterilization -- 10^{-4} probability of survival of one organism -- must apply equally to the interiors of the components. The fact that only a small percentage of the internal contaminants may be released into the environment has no logical bearing on this proposition, since the release of only one microbe is theoretically sufficient to contaminate the planet.

Dry Heat Sterilization of Interiors of Components 6.

Only two sterilization methods are known to penetrate all solids: Heat and radiation. The technology of dry heat sterili-

zation of spacecraft surfaces and components is rather well advanced owing to experience with the Ranger lunar probes. Methods and effectiveness of dry heat sterilization of interiors or components are still, however, essentially unknown owing not only to lack of experience but also to the extreme difficulty of determining when and if the internal contaminants have, in fact, been destroyed. **The** currently recommended heat sterilization procedure, 135⁰ C. for 24 hours, was established on the basis of tests on the temperaturetime rate necessary to destroy what were considered the most resistant organisms encapsulated in soil. It is well known that clean organisms are more easily killed than dirty ones. It has been assumed that the protection afforded by soil particles is comparable to that given by the encapsulating spacecraft component material. This assumption is reasonable, but it remains unproven. Accordingly, the recommended rates for sterilization may be too low or unnecessarily high. The determination of an accurate, effective rate is desirable not simply as an academic exercise; it has a profound, at times decisive, effect on spacecraft design, materials, and instrumentation, for only those components which are heat resistant within required parameters can be used. The Conference stressed, therefore, the importance of further research needed to furnish definitive information on the kinetics of thermal destruction of encapsulated spaces. On the basis of existing information, the Conference considered that terminal

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sterilization by dry heat soak of the entire spacecraft assembly would be the preferable method of sterilization.

7. Sterilization by Radiation

The possibility of sterilization by ionizing radiation was briefly discussed. Extensive literature on the subject has indicated that it is not a practicable method to sterilize the entire probe since the radiation doses necessary to destroy all internal contaminants would impair the functioning or reliability of some components. Radiation may, however, be the method of choice for certain other components.

8. Sterilization by Other Techniques

Other possible methods of sterilizing the interiors of components were discussed. Fluid sterilants, such as formaldehyde, were considered of limited application owing to their propensity to damage or affect the properties of some materials. Proposals to include sterilants in the composition of solids were received with reserve for the same reason. The Conference believed, however, that the number of contaminants within the solids could be substantially reduced if manufacturing control were improved; the goal, "start clean and stay clean throughout the entire manufacturing process," was stressed.

9. Sterile Insertion

At the present time, certain essential components cannot

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withstand heat soak, the only sterilizing method considered feasible for terminal sterilization of the entire spacecraft assembly. To permit terminal sterilization of as much of the assembly as possible, it has been proposed that the components requiring special sterilization methods be inserted afterward using sterile insertion techniques.

The Conference was not in agreement concerning the practicality of sterile insertion. Some felt that to assemble a spacecraft in such a way that some parts could be added later, and to maintain terminal sterility under such conditions, could present impossible problems. All agreed that the ultimate objective should be, rather, to design components which do not require special handling. The development of sterile insertion techniques nevertheless was considered an important backup approach in the event that such components do not become available. In order to avoid planetary contamination, it is desirable to develop several alternatives. Otherwise, the dilemma may one day have to be faced of postponing a mission or using a contaminated probe.

Summary of Findings and Recommendations

- $1.$ The Conference endorsed the existing policy of the National Aeronautics and Space Administration to protect the planets, especially Mars, from contamination by terrestrial organisms transported by non-sterile spacecraft. It was noted that the levels of protection recommended by COSPAR appear reasonable and scientifically defensible as interim objectives, and should be followed until new information warrants their revision.
- $2.$ The Conference found that microbial contaminants in the interior of spacecraft components would constitute a hazard to planetary sterility if released on the surface. It is very difficult to evaluate the degree of this hazard owing to the serious deficiency of information in several key areas: The number and distribution of contaminants within solids, the longevity of the surviving organisms, and the probability of their release on the planetary surface.
- In view of this lack of basic information, the Conference strongly $3.$ recommended that research pertaining to the assay of encapsulated organisms be increased. With the development of reliable assay methods, the likelihood of planetary contamination from this source can be put on an experimental rather than a conjectural This will be the key to future progress. basis.
- The Conference could not justify relaxation of sterilization $4.$

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standards on the ground that encapsulated organisms are, by virtue of their entrapment, substantially less of a threat to the sterility of a planet.

- The Conference did not feel that the Martian freeze-thaw cycle $5.$ could be relied upon to destroy viable internal contaminants released on the planetary surface.
- The Conference considered terminal sterilization of the entire $6.$ spacecraft assembly by dry heat soak the method to be preferred on the basis of existing information. To this end, it noted the importance of developing easily sterilizable components.
- 7. In the event that terminal sterilization is not feasible, the Conference recommended that assembly of sterile components using sterile insertion techniques should be developed as backup technique.
- The Conference noted that sterilization by ionizing radiation 8. of the whole spacecraft is not feasible. It may, nevertheless, be the method of choice for certain components in the event that sterile insertion techniques are required.
- 9. Regardless of the seriousness of the special hazard to be associated with encapsulated contaminants, the Conference endorsed the principle of starting production with components

having the lowest achievable levels of contamination and of maintaining rigorous standards of cleanliness throughout spacecraft assembly.

 $10.$ The Conference favored a reconsideration of the special hazard posed by encapsulated contaminants at a future date, when more definite quantitative information has become available.

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