

THE DESIGN OF MARTIAN BIOLOGICAL EXPERIMENTS

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It is generally acknowledged that the search for extraterrestrial life is the most important scientific objective of space exploration. This proposition is so widely accepted that one might think that the discovery of life on Mars would immediately yield some important benefit to science and the human race. Such a discovery would be exciting, of course, and profoundly moving. It would be a masterpiece of technological achievement. But for science, the detection of life on another planet will not be an end, but only the beginning of a long series of investigations.

One of the fundamental questions we would ask about an extraterrestrial species is whether it originated independently of terrestrial life. It is currently a widely held view that life will originate in the universe wherever conditions are favorable--for example, wherever conditions resembling those of the primitive earth exist. Since conditions on primitive Mars were probably similar to those on the earth, this theory implies that the existence of an indigenous Martian biota is highly probable. If we wish to test this hypothesis, it is not sufficient to establish that life of some kind exists on Mars. It must also be shown that this life had a separate origin from life on the earth. This can only be done by comparing the chemical structures of the two forms of life. Fundamental differences in organization would indicate separate origins. If the two kinds of life are similar, the possibility that they had a common origin will have to be examined.

The discovery and preliminary description of Martian life will be attempted by means of automatic devices which relay the information they obtain back to the earth. For detailed analyses, however, it will probably be necessary to acquire Martian specimens for examination in terrestrial laboratories. The acquisition of such specimens should therefore be regarded as the ultimate goal of Martian biological studies (1).

In planning Martian biological experiments, it is essential to consider the forms that an indigenous Martian biochemistry would be likely to take. Speculative though they may be, such considerations are unavoidable, for example, in designing a medium for the Gulliver life-detection experiment described in this Symposium by Levin (2). What labelled compounds should be included in the Gulliver medium in order to maximize the probability of detecting Martian life? The suggestion that all organic compounds of possible biological significance be included is impractical. Even if such a medium were attainable it would be self-defeating, since experience shows that it would be more likely to inhibit metabolism than to support it.

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The Gulliver experiment and all others designed to detect metabolism assume that if life exists on Mars it will be carbonaceous. This is not solely because life as we know it is based on carbon, although this circumstance is not without significance. Silicon is the element usually referred to in speculations about non-carbonaceous life. When one considers the fact that silicon is far more abundant on the earth than carbon, it may seem strange that it plays an almost insignificant role in terrestrial biochemistry. The reasons for this become clear, however, when one examines the properties of silicon compounds. First, they are unstable, owing to the weak bonding energy of the Si atom. The Si-Si bond has about half the energy of the C-C bond (3). Even such a simple compound as disilane, Si_2H_6 , the silicon analog of ethane, decomposes at moderate temperatures. Second, compounds of silicon are exceedingly reactive. In contrast to the chemical inertness of the hydrocarbons, most of the hydrides of silicon are spontaneously inflammable in air, and they are decomposed by water. Third, silicon dioxide, the end product of many reactions of silicon, is unlike carbon dioxide in being insoluble, involatile, and inert. These properties of silicon dioxide are explained by the fact that silicon has little tendency to form double bonds (4). The same properties that disqualify silicon for a significant biological role on the earth would do the same on Mars.

If carbon is uniquely suited for the construction of biologically useful substances, what can be said about carbon compounds? Is there a select group of these which are uniquely fitted for biological functions? A priori, this would seem to be unlikely, yet the chemical composition of living matter is, if anything, remarkable for its monotony. All species that have been examined, from viruses to man, make their genes from nucleic acid and their enzymes from protein. With a few minor variations, these are always built out of the same 8 or 10 nucleotides and 20 amino acids. To the extent that the characteristics of living things are determined by their genes and enzymes, it can be said that there is only one form of life on the earth, and this is composed of a surprisingly small number of primary organic compounds.

This uniformity of chemical composition is the strongest evidence we have for the common origin of known species. At the same time, it raises some questions that are relevant to the search for life on Mars. For example, why, out of the large number of possible amino acids, was the particular set of 20 selected that is found in proteins? Is there a fundamental reason for this selection, or is it a matter of chance? Probably both, to some extent. The experiments of Miller (5) have shown that α -amino acids are prominent products of organic syntheses in a simulated primitive atmosphere, and among these are several of the amino acids commonly found in proteins. Still others are obtained when the experimental conditions are varied (6). It has recently been shown by Ponnampertuma *et al.* that adenine, an important constituent of nucleic acids, is also produced by irradiation of a simulated primitive atmosphere (7), a result that had been foreshadowed by the work of Oro (8). The same workers have identified ribose and deoxyribose in heated or irradiated solutions of formaldehyde and acetaldehyde, substances known to be formed in the Miller experiment (5). Despite the fact that some of the interpretations are open to criticism (9), it now appears likely that the synthesis of most, if not all, of the building blocks of proteins and nucleic acids will eventually be accomplished under plausibly

"primitive" conditions. The choice of these substances for the formation of living matter may thus have been dictated at least in part by their availability.

One implication of this argument is that if other kinds of substances had been available, they also might have been used biologically. This raises a difficult problem, since there is reason to suppose that other substances suitable for protein and nucleic acid synthesis were, in fact, available on the primitive earth. To take one example, α -aminobutyric acid is a product of the Miller experiment (5). If it was present on the primitive earth, there is no obvious reason why it should not have been utilized for protein synthesis. It is a common metabolic product of plants and animals, yet it is not found in proteins. This suggests that chance may have played a part in the selection of amino acids for protein synthesis, just as chance, in all probability, determined which of the two optical forms of the amino acids would be used (9).

Another relevant question is the following: Given that a certain set of amino acids was selected for one reason or another for incorporation into the proteins of primitive organisms, how can we explain the persistence of this set, unaltered, over the long course of evolution? It is a remarkable fact that, with a few exceptions which I will return to, proteins of the most diverse functions and from the most diverse species are composed of precisely the same 20 amino acids, although a much larger number are formed in metabolism. At least 100 amino acids have been identified in plant extracts, and the number is still increasing (10). Does this mean that the 20 common amino acids are uniquely suited for building enzymes? This is dubious on general grounds, and there is some evidence against it. Amino acid analogs can be incorporated into proteins, and, in some cases at least, incorporation of an analog does not hinder the normal functioning of enzymes (11). Of course, it is one thing to ask whether an analog can replace an amino acid in a protein which is otherwise normal in constitution, and another to ask whether an entirely new set of amino acids capable of forming active enzyme molecules is possible. Since the 20 common amino acids have been selected for mutual compatibility, it is not likely that a random change in any one of them will be tolerated. On the other hand, there is no reason to doubt that other compatible sets exist.

Probably the strongest force opposing evolutionary changes in the amino acid set is to be found in the mechanism of protein synthesis. This mechanism, as currently understood (12), is outlined below:

1. DNA + Ribonucleoside triphosphates $\xrightarrow[\text{polymerase}]{\text{RNA}}$ m-RNA
2. Amino acid (AA) + ATP + s-RNA $\xrightarrow[\text{enzyme}]{\text{Activating}}$ s-RNA-AA + AMP + PP
3. Ribosomes + m-RNA + n(s-RNA-AA) $\xrightarrow[\text{Enzyme(?)}]{\text{GTP +}}$ (AA)_n + n(s-RNA) + m-RNA

The incorporation of an amino acid into protein, (AA)_n, requires the participation of a specific activating enzyme, a specific transfer-RNA

(s-RNA), and a specific triplet of bases in "messenger"-RNA (m-RNA). The enzyme activates the amino acid and attaches it to s-RNA. This goes to the ribosome, where it combines with the corresponding coding triplet in m-RNA. In due course, the amino acid is added to the end of the growing polypeptide chain. This mechanism appears to have great evolutionary stability--i.e., great resistance to changes in the number or kinds of amino acids in its repertory--because in order to effect such changes, coordinate mutations must occur in many elements of the system.

The amino acids hydroxyproline and hydroxylysine are of special interest in this connection. These are not among the universal twenty, yet they occur in collagen, a structural protein that is found throughout the animal kingdom. Hydroxyproline also occurs in the cell-wall protein of plants. It has been found that neither of these hydroxyamino acids is recognized by the amino acid activating enzymes, and consequently neither of them can be incorporated into collagen as such. Instead, they are formed from proline and lysine after these have been activated by their specific enzymes. Thus, the hydroxy acids are sneaked into the protein by a subterfuge (13,14).

In summary the following conclusions seem to be justified:

First, Martian life, if it exists, will be carbonaceous.

Second, it is likely to be similar, but not necessarily identical, to terrestrial life in chemical composition. This follows from the assumptions that the primitive Martian atmosphere resembled that of the primitive earth, that similar chemical reactions occurred in it, and that Martian life, like terrestrial life, was formed from the compounds available.

And third, the substrates most likely to be metabolized by Martian life are simple organic compounds like those produced by the irradiation of simulated primitive atmospheres.

If these conclusions are correct, then the discovery of Martian life, if it exists, will not be too difficult a task for our automatic devices; but the determination of whether and to what extent it differs from terrestrial life in its fundamental chemistry could be a difficult problem indeed.

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