

MULTIVATOR

Proposal for Mariner B Experiment - Capsule

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Submitted by

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The multivator is a miniature, multiple purpose "laboratory" in which a series of simple measurements can be made on samples of atmospheric dust. A variety of measurements are proposed, and others are to be considered. They have the common feature of testing a small sample of dust with a fluid reagent, and of giving a read-out by simple optical or electrometric measurement. In addition to a determination of solution or suspension properties (turbidity, pH, conductivity) the estimation of phosphatase and other enzymes for which ultramicro-tests are available would be of great help in estimating the possible existence of life on Mars.

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The effectiveness of the multivator depends on 1) the acquisition of samples of surface material from atmospheric dust or from the ground, and 2) the choice of feasible but informative and sensitive tests to conduct on these samples. Part one will be the special responsibility of the development group at JPL. Part two will be the preoccupation of the Exobiology Laboratory at Stanford University. Our continuing studies are based on the assumption that several samples of about one milligram of surface material will be available ("Clean" terrestrial air contains ~ one milligram dust per $10M^3$: a 100 km column of 1 cm cross section). Plainly this assumption would most readily be satisfied with access to the ground environment; however, Mars' atmosphere may be dustier than the Earth's. Of the biologically oriented experiments proposed hereinbelow, the determination of phosphatase with the use of substrates that release a fluorescent chromogen appears to be the most promising gamble for the detection of evidence of life.

We propose a device containing 24 test chambers (two circular plates with 12 each) containing about one ml of fluid each and into which the dust sample can be introduced. The plate will be rotated on its axle to allow each chamber in turn to intercept a light beam for photometric measurements. Other chambers will have built-in electrodes for measurements of electric conductivity or potential. The JPL will assume responsibility for and has begun preliminary work on the problems of mechanical design of the multivator and the collection of the dust. The Exobiology laboratory at Stanford will conduct the calibration of suitable test reactions and attempt to develop more sensitive and reliable assays.

The following measurements are now proposed or under review to be

conducted on aqueous suspensions of the dust samples. Refined variants are also indicated. In some cases, the same chamber can be used for several parallel measurements.

A. Solution Properties

1. Turbidity - for rough calibration of further experiments
2. pH
3. Conductivity
 - α - polarographic measurement (help to identify any electrolytes).

B. Enzyme Tests

1. Phosphatase - colorimetric test by splitting of p-nitrophenyl phosphate to release nitrophenol
 - α - fluorimetric test by splitting of fluorescent phosphates
2. Deoxyribonuclease - conductimetric test by release of dialyzable nucleotides from DNA
 - colorimetric test by splitting of p-nitrophenyl-thymidine-phosphate
3. Ribonuclease - tests analogous to β₂ with RNA substrates
4. Other esterases (sulphatase, acylase) and glycosidases by methods analogous to 4.

C.

1. Electron transfer enzymes
2. Substrates for electron transfer enzymes
3. Microbial growth on defined media by changes in turbidity, conductivity or pH of the biochemical tests.

Phosphatases (β₄) are among the most ubiquitous enzymes - I know of no tissue or organism that has failed to show them - and they can be readily demonstrated in small samples of soil, dust, sediment from tap water, sea water (about 3×10^{-9} moles of nitrophenol released per milligram soil per hour). This

is just within comfortable detectivity in a laboratory spectrophotometer, and might also be achieved by differential colorimetry in a compact device. For example, a dual light source might consist of two pin lamps with differential filters, and driven by AC modulated in opposite phase. Differential absorption of one color would be recognized as an AC output from the detector.

This list is oriented to the detection of clues to life. In addition, the measures of part A, together with other microscopic and video reconnaissance studies will be invaluable preparation for the more ambitious efforts of the Voyager series.

We are studying the extension of these microtests for phosphatase and other enzymes as well as possible artifacts and improvements. The presence of phosphatase activity in soil would be presumptive evidence of life related to the utilization of phosphate and can establish the limits of its prevalence.

The sensitivity of this test can be augmented by the use of fluorescent substrates. Dr. Rotman (now working in my laboratory) has shown that the detectivity with these substrates is several orders of magnitude higher than with nitrophenyl compounds and he can readily demonstrate the enzyme level of single bacteria, perhaps even single enzyme molecules.

Equally important and widespread enzymes are the nucleases. We are studying ultramicro methods for these. One possibility is to contain the reaction mixture in a dialysis sac within the multivator chamber, with a film of distilled water in contact with probe electrodes. The release of low molecular weight, dialyzable nucleotides would be signalled by an increased electrolytic conductivity between the electrodes. Whether this can be made sensitive enough depends partly on the electrolyte content of the soil itself.

The instrumental aspects of this proposal can be discussed by JPL. As a preliminary estimate, the design would cost about 5 lbs (mostly the dust collection which might be integrated into the structure of the capsule) and <5 watts power. The information output would be of the order of 1-10 bits per second. We are considering further ways of minimizing the weight of the package. The reagents themselves would, of course, weigh only an ounce altogether; and the signal outputs are as simple as from any transducer.

It would, of course, be foolish to delay too long the final decision on

test reactions and the ways these can be incorporated into the multivator. However, the studies on ultramicro assay methods can proceed in parallel with the mechanical and electronic design of the multivator since the basic output from the chamber will be the same regardless of the type of reagent used for the individual test and of the choice between a photometric or more direct electro-metric estimation.

The work at Stanford is already adequately funded as an aspect of our current investigations and we are adding additional staff during the next two or three months to help to accelerate them. Additional support may well be required for the instrument development work at JPL. It should be possible to construct at least an elementary version of the multivator with tests and controls for, say, items A, 1, 2, 3 and B1, within the indicated schedule.