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A Simulation of the Viking Labeled Release Experiment for a Nonmajors Astronomy Course

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Abstract

This article describes a laboratory exercise that demonstrates some of the principles behind the Viking Labeled Release experiment. It is suitable in complexity and depth for a nonscience majors astronomy course.

1. INTRODUCTION

One of the major challenges in designing astrobiology experiments for the undergraduate teaching laboratory is providing an experience for the student that is authentic and meaningful and that conveys one or more basic principles of astrobiology, but that is relatively straightforward in implementation. This is particularly difficult in the case of introductory astronomy courses for nonscience majors. In these courses, students often have little or no science education beyond the secondary level. To compound the problem, most instructors in such courses are (naturally) astronomers whose grounding in astrobiologically related fields such as chemistry and biology may be minimal.

The following is a description of a laboratory exercise that simulates one of the major experiments in the history of astrobiology: the Viking Labeled Release experiment. This exercise can be performed in almost any setting, classroom or laboratory. The only utility requirements are hot and cold running water. None of the components of the experiment is toxic, flammable, or corrosive.

2. THE VIKING BIOLOGY EXPERIMENTS

In 1976, the Viking mission landed two spacecraft on the surface of Mars. Each of these landers carried a package of three experiments designed to look for the by-products of microbial metabolism in Martian soil (Brown et al. 1978). This was done by adding a nutrient mixture to soil samples in an enclosed chamber and monitoring the gases produced or consumed. One of the Viking biology experiments, the Labeled Release experiment (Levin 1972), used radioactive carbon-14 in the nutrient mixture to detect production of carbon dioxide (CO₂) from biological respiration.

One of the characteristics of living microorganisms is that their growth and metabolism follow a distinct pattern when the cells are provided with a fresh supply of nutrients. This growth curve typically has three phases: a lag phase, in which the cells are synthesizing enzymes and metabolic intermediates necessary for growth; a growth phase, in which the cells are dividing exponentially; and a stationary phase, when the cells have reached a population size that is limited by available nutrients and energy or the buildup of waste products. The production of metabolic by-products, such as CO_2 , will also follow this pattern of lag period followed by an exponential increase in concentration. The Viking Labeled Release experiment was designed to detect microbial metabolism by measuring the release of CO_2 .

Nonbiological chemical reactions, on the other hand, usually start as soon as the reactants are brought together, and continue at a constant rate (i.e., a linear increase in concentration with time), determined by environmental conditions, until the reactants are completely consumed. This difference in behavior between biochemical and abiotic chemical reactions can be used to distinguish between the two types of processes if the extent of the reaction is measured over time. Although the Viking Labeled Release experiment and the other Viking biology experiments were designed to detect metabolic by-products with little thought of possible interference from chemical reactions (Klein et al. 1976; Klein 1979), the failure of the gas chromatograph/mass spectrometer to detect even small traces of organic material in the Martian soil led the investigators to conclude that oxidative chemistry, rather than biology, had generated the CO₂ measured by the Labeled Release experiment (Klein 1978).

3. THE LABORATORY EXERCISE

The laboratory exercise described below simulates the Viking Labeled Release experiment. The procedure has been adapted from Corton et al. (2002). "Soil" samples are prepared by adding to sand either dried baker's yeast (to produce a biological source of CO_2) or sodium bicarbonate and potassium hydrogen phthalate (to produce a chemical source). A "nutrient solution" of sugar and water is added to each sample, and the evolution of CO_2 is measured as a function of time. Corton et al. used a CO_2 -sensing electrode and had the students plot voltage versus time to obtain the reaction curve for each sample. Although this approach is quite appropriate for a chemistry class, it may be too complex and equipment-intensive for use in a nonmajors astronomy or "life in the universe" course. The modification described here uses a pH indicator dye, phenol red, that is placed in a test tube connected to the reaction flask by rubber tubing. This dye changes color from red to yellow when CO_2 diffuses from the sample, down the tubing, and into the indicator solution, where it dissolves in water and lowers the pH to around 6. The students measure the amount of time that the color change takes for each sample, giving them a more qualitative but still valid demonstration of the principles used by the Viking biology experiments.

4. SET-UP

Students should be divided into at least three groups. The following equipment is needed for each group:

- 1 125-ml Erlenmeyer filter flask (with side arm)
- 1 #5 rubber stopper
- 1 piece of rubber or Tygon tubing, i.d. approx. 0.7 cm, length approx. 15 cm, with tape wrapped around one end to fit snugly into the mouth of the test tube
- 1 test tube, approx. 1.25 cm i.d. by 10 cm length (vol. approx. 10 ml), with 1 ml level marked on the side of the tube
- 1 plastic food storage tub, approx. 20 x 12 x 6 cm
- 1 small plastic funnel

The following solutions should be prepared before class:

Nutrient solution: 1 part sucrose or glucose to 4 parts water, 20 ml per group. Indicator solution: approx. 0.1 mg phenol red per ml water, 1 ml per group.

The following "Martian soil" samples should also be prepared before lab:

Biological sample: Approx. 1/3 package dry baker's yeast in 50 cc sand.

Chemical sample: Approx. 5 g each sodium bicarbonate and potassium hydrogen phthalate in 50 cc sand. Control or Sterile sample: 50 cc sand.

Label these samples as unknowns (e.g. "A," "B," "C") and place in plastic sandwich bags. If more than three groups are to participate, duplicate samples should be prepared. The sand should be clean, purchased prepackaged from a garden store if possible. The sodium bicarbonate and potassium hydrogen phthalate can be purchased from Sigma Chemical Company or other chemical suppliers. A source of warm tap water should be available nearby.



Figure 1. The entire experimental setup at the beginning of the exercise



Figure 2. An assembled apparatus for one group

5. STUDENT INSTRUCTIONS

- 1. Connect the untaped end of the plastic tubing to the side arm of the flask.
- 2. Obtain your soil sample from the instructor. Note the label on the sample you are given. Pour the soil sample into the flask using the funnel.
- 3. Fill the test tube with indicator solution up to the line marked on the tube. Connect the test tube to the flask by placing the taped end of the plastic tubing into the mouth of the test tube.
- 4. Fill the tub about 1/2 full with warm tap water and set the flask in the tub.
- 5. Pour the nutrient solution into the flask and stopper the flask. Begin timing.
- 6. Stop timing when the solution in the test tube has changed color from red to yellow, or when the instructor advises you to stop.

6. EXPECTED RESULTS AND DISCUSSION POINTS

Using the concentrations of solutions and amounts of ingredients given above, the "chemical" sample should begin to produce CO_2 immediately, and should turn the indicator solution yellow in 5 to 10 minutes. The "biological" sample should begin to produce CO_2 after approximately 10 minutes, and should turn the indicator solution yellow in 20 to 30 minutes. These times will vary somewhat if different

concentrations of solutions and ingredients are used, or if flasks and tubes with different volumes are used. The students will probably observe the formation of bubbles in the solution before any color change is seen in the indicator dye. This observation can be used to point out that the Viking biology experiments as flown had no method of visual observation of the samples during the experiments, although a light-scattering experiment to measure cell division was originally part of the package and was omitted from the final flight version (Klein 1992).

The initial report of data from the Viking Labeled Release experiment can be found in Levin & Straat (1977). The interpretation of this experiment has remained somewhat controversial over the ensuing years. The consensus view in the scientific community remains that the Viking biology experiments found reactive chemical processes, but no life, in the Martian soil (see Klein 1978). Levin, however, has maintained his view that his experiment detected Martian life (see Levin & Straat 1981). A summary discussion of the Viking biology experiments can be found in Jakosky (1998).

Completely faithful reproduction of complex scientific experiments is not always possible in the classroom or teaching laboratory. This is particularly true if the experiment in question was performed by a spacecraft on another planet. In any classroom adaptation of an experiment, a balance must be struck between making the exercise logistically feasible in the target classroom environment and faithfully conveying the scientific principles on which the original experiment was based. In the Viking Labeled Release simulation described here, the target classroom environment is one in which the demands for facilities, equipment, and prior student experience must be kept to a minimum. This approach dictates a shift from the more complex experimental set-up of Corton et al. (2002), in which the evolution of CO_2 is monitored over time, to the approach used here in which the data consist of a single point in time at which the CO_2 concentration reaches a level sufficient to cause a color change in the indicator solution.

This design naturally limits the accuracy of the simulation to some degree, and this must be taken into account when using the exercise in the laboratory or classroom. The instructor should point out that in the laboratory exercise, we are assuming that the overall rate of CO_2 production from a chemical reaction will be faster than that from microbial metabolism, primarily because of the lag phase typical of microbial growth curves. Thus, the difference in time of color change between the two samples is assumed to represent only the presence or absence of a lag period before CO_2 production starts. This assumption may not be valid if, for example, the sample has a very low level of chemical reactant or a very high level of microorganisms. In fact, the implausibly high cell density required to explain the rate of CO_2 production observed on Mars was one of the factors that led to the chemical interpretation of the Labeled Release data (Klein 1978).

We can test this assumption to some extent by observing through the transparent walls of the flask the formation of gas bubbles (which are assumed to be CO_2) in the liquid, and noting that the chemical sample starts to form bubbles immediately upon addition of liquid, while the biological sample does not form visible bubbles for a few minutes. If the instructor deems it appropriate, the students could record the time of first visual bubbles and use this to further differentiate the behavior of the "biological" and "chemical" samples.

7. EXPERIENCE WITH THE EXERCISE

This laboratory exercise was used by the author in a nonmajors astronomy/life in the universe laboratory class at Pomona College in fall 2003. It was performed several times with classes averaging five to six students each. The Viking mission and the general topic of life on Mars had been covered previously in the lecture portion of the course. No formal evaluation of student material retention was attempted, but student reaction was generally good. The students were engaged, asked questions, and participated in discussions of several of the points mentioned above. The level of complexity of the exercise appeared to the author to be appropriate for a group of nonscience majors with good overall academic skills, but little or no previous exposure to science lecture or laboratory courses at the college level. The author welcomes feedback from other instructors who may use the exercise.

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ÆR 98 - 105