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## *News & Views*

# Comment on "Stereo-Specific Glucose Consumption May Be Used to Distinguish Between Chemical and Biological Reactivity on Mars: A Preliminary Test on Earth"

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Sun, H.J., Saccomanno, V, Hedlund, B., and McKay, C.P. ("Stereo-Specific Glucose Consumption May Be Used to Distinguish between Chemical and Biological Reactivity on Mars: A Preliminary Test on Earth") is an important and timely paper addressing the recently revived interest in the possibility of extant life on Mars. The paper specifically and succinctly proposes to resolve the 33-year aura of doubt surrounding the Viking Labeled Release (LR) life-detection experiment (Levin and Straat, 1976). Reporting on experiments performed on all three domains of life, archaea, bacteria and eukaryotes, Sun *et al*. convincingly show that chirality preference distinguishes metabolic consumption of substrates from purely physico-chemical reactions in that the latter have no chirality preference. The Sun group proposes their method to determine whether the Viking LR experiment's positive response was from living microorganisms (Levin, 1997) or from a physico-chemical agent in the martian soil (Klein, 1999).

Sun *et al.* used wet chemistry to determine microbial uptake of glucose from a racemic nutrient solution to demonstrate biological chiral preference, and a respirometer followed by a gas analyzer for $CO_2$ to demonstrate that the strong oxidant permanganate attacks both the L- and D-isomers of glucose equally. These complex analytical systems would be difficult to instrument for robotic astrobiological use. They are also orders of magnitude less sensitive than the LR radioisotope technique, which raises the question why the researchers did not use that legacy method.

The Viking LR addressed the chirality distinction between living and abiotic reactions, but spacecraft constraints prevented the use of separate instruments for the different chiral isomers. However, those isomeric compounds in the nutrient were included in racemic mixtures to avoid missing an alien life with a chirality different from ours. The Chiral LR miniature instrument was subsequently proposed (Levin, 1987) to identify each specific isomer in the nutrient that obtained a response from the soil. Since the Viking LR produced the response Sun *et al.* propose to investigate, it would seem important to include those LR chiral substrates, L- and D-lactate and L- and D-alanine, in any return to Mars for the stated purpose of resolving the origin of the Viking LR signals. Thus, it would have been desirable for the Sun group to have included those isomers.

There is a more fundamental problem. The method proposed would fail to detect $CH_4$ from methanogens, should they, as increasingly speculated (Krasnopolsky *et al.*, 2004; Mumma *et al.*, 2009), exist on Mars. On the other hand, the LR experiment would readily detect $CH_4$ emitted by the many methanogens that metabolize organic compounds (Escalante-Semerena and Wolf, 1984), including formate, one of the Viking LR nutrients. Strict lithotrophs could also be detected by using $^3H_2O$ as solvent for the LR nutrients instead of $H_2O$ and monitoring for $C^3H_4$ after it passes through a vapor-impermeable membrane or barrier (to exclude $^3H_2O(g)$ from the detector). In this manner, labeled gas evolved from any labeled substrate applied to the test soil can be detected by the LR instrument, as, for another example, $H_2S$ from $^{35}S$-labeled cysteine. No special wet chemistry analytical techniques and their respective instrumentations need be applied.

Another difficulty is the selection of glucose as the substrate of choice. Prevailing COSPAR requirements (COSPAR, 2002, 2005) prevent the use of glucose in life-detection missions because it cannot withstand the heat of sterilization prescribed. This position was recently supported for soil samples to be taken beneath the UV depth (the Viking LR samples were taken to a depth of 4 cm) by one of the paper's authors (McKay, 2009). It was for this reason that glucose, despite its strong metabolic attributes, was not included in the Viking LR.

In the final analysis, the paper makes a good case for chiral preference as a means to get a widely acceptable answer to the tantalizing question of what the Viking LR found in the surface soil of Mars and for carrying out future astrobiological investigations of detection and comparative biology between terrestrial life and any alien forms.

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## Abbreviation

LR, labeled release.

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# Response to Dr. Levin's Comment

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In his comment on our paper, "Stereo-Specific Glucose Consumption May Be Used to Distinguish between Chemical and Biological Reactivity on Mars: A Preliminary Test on Earth," Dr. Levin raises several important points on how to improve a chiral life-detection experiment on Mars. Levin suggests that the radioactive label is the method of choice on space missions. We agree, even though the method is less practical for routine laboratory use here on Earth. He also suggests that the use of D- and L-lactate and D- and L-alanine be considered. We agree, and that work is in progress. The issue of detection of methanogens is interesting and opens up the question of how to devise the optimal set of chiral molecules in a life-detection experiment. This puzzle is not yet solved, but careful experiments with a variety of compounds on a variety of organisms are needed. Levin's suggestions for using labeled water and labeled sulfur in cysteine are novel and should be considered as the instrument design moves forward.

The question of spacecraft sterilization spans both scientific and regulatory issues that must be solved as flight instruments are designed. Our goal at this point is to accumulate a body of empirical data on what chiral substrates may be used for a flight instrument. Later, these results can be folded into an instrument design along with other engineering and regulatory requirements.

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