It's life… isn't it?

Scientists find it hard enough to pin down evidence of early life on our own planet. How on Earth do we plan to determine whether life exists elsewhere? John Whitfield finds out.

Where can, it's fair to say, be confident that there's life on Earth — but proving it is a different matter. In December 1990, the Galileo spacecraft pointed its sensors back at Earth before setting off for Jupiter. The probe reported an atmosphere abundant in oxygen and unusually rich in methane. It also detected a mysterious pigment that was unlikely to be of mineral origin: something earthlings call chlorophyll. Yet the astrophysicist Carl Sagan and his colleagues were still cautious in their conclusions based on these results. "Together, these are strongly suggestive of life on Earth," they wrote¹.

Galileo, admittedly, was not designed to detect life. And it did have a rather distant view of our planet, with only hints from the atmosphere to work out what was going on at the surface. But some dedicated, close-up examinations have proven no more conclusive. Efforts to detect faint traces of life in Earth's oldest rocks, some 4 billion years old, and in martian rocks that have fallen to Earth, have likewise produced results that are both ambiguous and disputed; claims of errors in procedure and interpretation fly back and forth with regularity.

Nevertheless, space agencies are shifting

their research agendas towards looking for life, says chemist Richard Mathies of the University of California, Berkeley. "Exobiology is becoming more prominent," he says. NASA and the European Space Agency (ESA) are moving from rock and mineral experiments to biological ones, he explains. In 2009, both agencies will send landers to Mars that, unlike the Spirit and Opportunity rovers now on the planet, will be designed to look for the chemistry of life.

Astrobiologists are currently working out what to look for, designing the instruments that could detect it, and using the most inhospitable, Mars-like environments on Earth as training grounds. Even so, these missions are highly unlikely to give a definitive answer. For that, we will have to wait at least another decade, when planned missions will bring back martian rocks for study in terrestrial laboratories.

The first experiments to look for life on Mars were the two Viking landers of 1976. Each lander scooped up a spoonful of soil from the planet's surface, moistened it with pure, sterile water, incubated it, and watched for evidence of chemical processes similar to those found in terrestrial microbes — such as the uptake of nutrients or exchange of gases. As a control, the landers did the same tests on a heat-sterilized sample of martian soil.

Remarkably, every test gave a positive result. The soil samples released oxygen and compounds made from ingredients in the nutrient solution, and carbon in the experiment seemed to be incorporated into organic molecules. Unfortunately, most of the controls — except the experiments designed to detect nutrient uptake — also gave positive results. At the same time, two instruments designed to analyse the planet's chemistry, a gas chromatograph and a mass spectrometer, failed to detect any organic compounds on the surface of Mars².

Hit and miss

In sum, these experiments gave us some clues about the chemical environment on Mars, but failed to confirm or refute the existence of life. "The experiments all turned out to have some element of ambiguity, but that's utterly understandable," says chemist John Kerridge of the University of California, San Diego. Most researchers conclude that inorganic chemical reactions, perhaps powered by ultraviolet light, must have produced the chemicals detected³. A few still argue that the positive

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results should be taken as evidence for living microbes.

Ambiguity wasn't the study's only problem. Even if Viking had delivered a definite 'no' to life, this could simply have been because its instruments were too crude. "There could have been 10 million bacteria per gram of martian soil, and Viking wouldn't have seen them," says Andrew Steele of the NASA Astrobiology Institute at the Carnegie Institution of Washington.

Facing such problems, interest in looking for life on Mars waned after Viking, says astrobiologist Christopher McKay of the NASA Ames Research Center in Moffett Field, California. "The experiments were very hard to follow up on," he says. But there has been a boom in instrumentation since then - today's detectors can spot a single cell, or molecules at concentrations of parts per trillion. And our understanding of the tenacity of life in extreme environments has grown. On Earth, evidence for bacterial life has been found everywhere from deep oceans devoid of light, to hot springs at temperatures above boiling point. By the mid-1990s, biologists were once more on a mission to determine whether life could exist elsewhere in the Solar System — particularly on Mars, Jupiter's

Spot the difference: the Atacama Desert (above) is very similar to the martian landscape (left), which makes it an ideal place to test equipment for probing Mars for signs of life.

moon Europa, or Saturn's moon Titan.

Astrobiologists still face the problem highlighted by Viking: how do you ensure that 'evidence' of life isn't something produced by non-living processes? Such concerns have been at the heart of the debate on ALH84001,a 4.5-billion-year-old martian meteorite found in Antarctica in 1984. A team led by David McKay of NASA's Johnson Space Center in Houston, Texas, argued in 1996 that several features of the rock might have been produced by living organisms⁴. These were organic compounds, minerals similar to those produced by Earth bacteria, and structures resembling fossil bacteria.

But in the years following the publication, these claims came under sustained criticism. Some features, such as the proposed microfossils and mineral structures, could have been produced by non-

living chemistry^{5,6}. Others, such as the organic chemicals present in the rock, may have been added to it after it arrived on Earth⁷. Few researchers now believe that

ALH84001 is good evidence for the presence of past life on Mars.

Yet even those who disagree with the initial claims made for ALH84001 are glad of the effect the study had on the field. "Exobiology came out of the closet with that paper," says Mathies. "Suddenly it was OK to be working on this stuff." Christopher McKay and his colleagues also helped to define the categories of evidence — organic chemicals, minerals and fossils — necessary to build a convincing case for martian life, says Steele. "Even if the answers were wrong, the approach was good — they raised the bar by several metres."

Similar, and sometimes acrimonious, debates swirl around the evidence for the oldest life on Earth. Whether or not an outcrop of rock in Greenland contains evidence of life from 3.8 billion years ago is hotly contested⁸. And the question of whether structures in

"We've not yet identified a smoking gun for life." — Jack Farmer

3.5-billion-year-old rocks in Australia are fossilized bacteria or artefacts produced in hot springs has been controversial for several years^{9,10}. Those astrobiologists not actively involved in early-Earth research keep an eye on its developments, hoping that it can provide a blueprint for their field. "The issue of whether these microfossils have a biological origin or not is so contentious; it really shows the need to attack the problem from as many angles as possible," says Jorge Vago, study scientist on ESA's 2009 ExoMars project.

Both morphological and chemical signals are needed to confirm a claim. But both have their problems. "We've not yet identified a smoking gun for life," says geologist Jack Farmer of Arizona State University, Tempe.

A rocky road

The most recent attempt to look for evidence of life on Mars, the ill-fated Beagle 2 lander that presumably crashed on the planet last Christmas, was equipped to dig into the martian soil, to escape the surface environment that destroys organic compounds, and study the isotopic ratio of different elements. On Earth, living things preferentially incorporate the lighter form of carbon. The ratios of different isotopes of sulphur, iron, nickel and chromium have also been proposed as signs of biological activity.

But, again, it is not clear how geology and chemistry influence isotope ratios, particularly in rocks that may be billions of years old. Some believe that the carbon in Greenland's ancient rocks has a volcanic, rather than a biological origin^{11,12}. Geological processes that transform carbonate minerals into organic matter seem to produce compounds that, like biological remains, are enriched with the lighter form of carbon, says geo-

chemist Mark van Zuilen of the Petrographic and Geochemical Research Centre in Nancy, France. "I don't think that carbon isotope ratios are that definitive an indicator of life," he says.

"We probably don't know all the ways in which isotope fractions can come about," adds geologist John Parnell of the University of Aberdeen, UK. "There may be things going on that we haven't thought of yet." Parnell is leading a UK project to identify biological molecules on the early Earth and Mars. His favoured biomarker is a class of organic molecules called hopanes, which are produced from the breakdown of simple cell walls. They are very long-lived — on Earth they persist for billions of years — and we know of no inorganic process that can produce them.

Mathies and his colleagues, on the other hand, believe that a good starting point is to look for amino acids. "If you look at life on Earth, 50% of the mass of biological materials is amino acids," he points out. These molecules can be made by inorganic processes as well as organic ones, and they have been

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found on comets and meteorites, which might confuse matters. But it is important not to be too ambitious at first, says Mathies: after going to the expense of getting to Mars, you want to give yourself a reasonable chance of finding something. "You want to start general and get more specific," he says.

And there may be more telling evidence available from amino acids — on Earth, all biological amino acids have a certain geometry, whereas those not made by organisms are evenly split between two mirror-image conformations. In space, a bias either way could be a good indicator of biology at work. Amino-acid detectors are also more sensitive than isotope measurements, says Mathies. His team has proposed an instrument for NASA's 2009 Mars Science Laboratory (MSL) project that could pick this up.

Other biomolecules that might be worth looking for in space include nucleic acids and sugars. Most researchers favour a search strategy based on the chemistry of terrestrial life, but going slightly broader. For example, life elsewhere might use different nucleotides or amino acids from life here, or favour a different geometry or bias of isotopes. But structure and patterns in any of these areas would all be pointers to life. "Complexity is the most interesting biomarker," says Steele.

Of course, no mission would limit itself to pursuing one particular line of investigation. Future Mars landers will brandish a range of different tests, aided by lab-on-achip technology that has made instruments much smaller, as well as more sensitive. The ExoMars lander, for example, will carry cameras and microscopes, subject rocks to spectroscopic and chromatographic analysis to gauge their chemical composition and, if they look promising, grind them up to look for amino acids and other organic chemicals. Animal, vegetable or mineral? Possible signs of ancient life in rocks in Greenland (above) and on the martian meteorite ALH84001 (inset) remain controversial.

It will also carry an antibody microarray chip that can detect extant life or contamination by microbes carried from Earth¹³.

NASA's effort will be slightly different. "The MSL is very much a chemical lab: it won't directly look for life, but maybe for the residues or precursors of life," says Firouz Naderi, head of the Mars programme at the agency's Jet Propulsion Laboratory in Pasadena, California. The MSL is a scouting party for the proposed 2013 Mars samplereturn mission and a 2016 astrobiology field laboratory; it may even hoard rocks for these landers to collect.

Out on the edge

To test all these sensitive instruments, researchers are currently digging through the dirt of Earth's harshest ecosystems. Parnell and his colleagues have been looking for organic chemicals in the Haughton impact crater, the relic of a past meteor impact in the Canadian Arctic, where NASA tests much of its Mars science and technology. Steele's team also spent last summer in the Arctic — this time in Svalbard, where the rocks share some geological features with ALH84001 — testing their life-spotting equipment on the local microbes. His team hopes to get a life-detection-chip on the ExoMars lander.

Christopher McKay and Mathies, mean-

while, are both working in the Atacama Desert in Chile. "It's the only place on Earth where there's no life at the surface at all," says McKay. He spent June looking for the residues of organic compounds on desert rocks, and digging holes to see if bacteria live underground there. The researchers work in special suits to avoid swamping any microbes in the soil with their own bacteria.

Another collaboration, between NASA and the Center for Astrobiology in Madrid, Spain, is looking for life, and testing the technology needed to drill and sample on Mars, in the acidic, mineral-rich Rio Tinto river in southern Spain. The acid is made by bacteria, which metabolize sulphur and produce sulphuric acid. Carol Stoker of NASA Ames, the project's leader, was delighted when the Opportunity rover landed on a plain of sulphate minerals on Mars that likewise seem to be the remains of an acidic sea. "It was a slam dunk," she says. "The chemistry of the martian surface is exactly like what Rio Tinto is producing now."

The researchers plan to test an automated drilling platform next spring, and also have an instrument, called SOLID (signs-of-life detector), that they hope to get on one of the 2009 missions.

So, despite the occasional false start and blind alley, the astrobiologist's tool kit and knowledge base is becoming ever broader. Most

researchers are now confident that, if there is or has been life elsewhere in the Solar System, we have the ability to detect it. But to do so, we will still have to be lucky enough to study the right rock in the right place with the right techniques — and get it safely back to Earth. "For a biosignature to be believed, it has to be returned for a sample," says Kerridge. "Reproducibility convinces hardnosed scientists." Even then, don't bet on everyone being convinced.

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- Sagan, C., Thompson, W. R., Carlson, R., Gurnett, D. & Hord, C. Nature 365, 715–721 (1993).
- Biemann, K. & Lavoie, J. M. J. Geophys. Res. B 84, 8385–8390 (1979).
- Plumb, R. C., Tantayanon, R., Libby, M. & Xu, W. W. Nature 338, 633–635 (1989).
- 4. McKay, D. S. et al. Science 273, 924-930 (1996).
- Bradley, J. P., Harvey, R. P. & McSween, H. Y. Jr Nature 390, 454 (1997).
- Barber, D. J. & Scott, E. R. D. Proc. Natl Acad. Sci. USA 99, 6556–6561 (2002).
- Jull, A. J. T., Courtney, C., Jeffrey, D. A. & Beck, J. W. Science 279, 366–369 (1998).
- 8. Dalton, R. Nature 429, 688 (2004).
- 9. Brasier, M. D. et al. Nature 416, 76-81 (2002).
- Schopf, J. W., Kudryavtsev, A. B., Agresti, D. G., Wdowiak, T. J. & Czaja, A. D. *Nature* **416**, 73–76 (2002).
- 11. Fedo, C. M. & Whitehouse, M. J. *Science* **296**, 1448–1452 (2002). 12. Van Zuilen, M. A., Lepland, A. & Arrhenius, G. *Nature* **418**,
- 627–630 (2002).
- 13. Clarke, T. Nature 413, 247-248 (2001).