



Cupriavidus metallidurans bacteria metabolize toxic gold chloride into gold nanoparticles (white).

MICROBIOLOGY

There's gold in them there bugs

Microbial 'alchemy' could lead to new ways of detecting and producing the precious metal.

BY PETER GWYNNE

Take a solution of gold chloride, a compound toxic to most forms of life. Add a colony of *Cupriavidus metallidurans*, one of the few bacteria able to survive amid compounds of heavy metals in mines across the world. As the bacteria accumulate the gold salt from the solution, biochemical processes within the organisms reduce it to the pure metal, which the bacteria excrete in the form of tiny gold nuggets — nanoparticles of pure gold. The bacteria produce the gold as protection from the toxic gold complexes

that would otherwise destroy their cells.

The process won't inspire a new gold rush, because scaling it up would be both prohibitively expensive and time-consuming, but it does have implications for the creation, detection and processing of this noble metal. And it all relies on extremophile microorganisms that can thrive in environments rich in solutions of salts that are lethal to most forms of life.

Recent findings may lead to bioindicators that can identify microorganisms associated with gold and hence pinpoint the presence of the metal; biosensors that can help prospecting teams quickly determine the concentration of

gold in environmental samples; and the biosynthesis of gold nanoparticles, which have potential applications in optoelectronics, imaging technology, catalysis, drug delivery and more. Researchers are also exploring whether bacteria played a role in the creation of gold deposits worldwide. Deeper understanding of the link between bacteria and gold could even lead to bacteria producing customized gold nuggets.

There are economic, environmental and scientific reasons for the current interest in the connection between bacteria and gold. The price of gold has risen by a factor of 4.5 during the past decade (see 'Mine, all mine!', page S2). This leap has made gold mining highly profitable. "But it also means that new deposits have to be found and difficult types of ore have to be processed," says Frank Reith, a geomicrobiologist at the University of Adelaide in Australia.

MICROBIAL ALCHEMISTS

Certain bacteria are "microbial alchemists, transforming gold from something that has no value to a solid, precious metal", according to Kazem Kashefi, a microbiologist and molecular geneticist at Michigan State University in East Lansing. "For millions of years they've been working on gold. They've had enough time to become specialists in gold production." Kashefi and his Michigan State University colleague Adam Brown, an expert in electronic art, have put that specialization to novel use in a work of microbial performance art. Their installation, called *The Great Work of the Metal Lover*, features *C. metallidurans* bacteria producing 24-carat gold from a solution of gold chloride in front of a live audience.

Today, researchers across the world are trying to find extremophiles that can produce pure gold from its compounds. The few types identified to date have been found on land, but researchers in India are searching the seas. "Marine water is known to be the richest source of gold in nature" in the form of gold chloride, says Anirban Roy Choudhury, a specialist in fermentation technology at India's Institute of Microbial Technology in Chandigarh. Choudhury leads a programme that studies the potential of marine microorganisms to create gold nanoparticles. Oceans also have extremely high temperature, pressure and salinity, and harbour heavy metal ions, all of which favour the growth of extremophiles.

Choudhury's team has isolated several types of bacteria from the seas around India and screened them for gold-producing ability. So far, he has found more than 15 species capable of producing gold nanoparticles. One microorganism, *Marinobacter pelagius*, proved particularly effective, rapidly synthesizing gold particles about 10 nanometres in size. The team is now trying to understand how the microbes produce gold nanoparticles. It also aims to develop what Choudhury calls "a suitable production and purification method for gold nanoparticles via biological routes".

FRANK REITH

Other teams are looking for gold-synthesizing bacteria on land. Geomicrobiologist Gordon Southam, director of the Centre for Environment and Sustainability at the University of Western Ontario in London, Canada, and geochemist Maggy Lengke, who was then a postdoctoral researcher on his team, found three types of bacteria with this ability: *Acidithiobacillus thiooxidans*, sulphate-reducing bacteria and cyanobacteria from a borehole more than three kilometres deep in the Driefontein Consolidated gold mine in South Africa's Witwatersrand Basin¹. "All three were able to produce solid gold from a gold solution," Lengke says.

Regarding the sulphate-reducing bacteria, "the bacterial precipitation of gold from the gold-thiosulphate solution is linked directly" to the microbes' ability to metabolize sulphur compounds, says Lengke. Such gold-containing solutions have been found in other gold sources, she notes, such as the Wau field in Papua New Guinea and the Ashanti mine in Ghana. The implication is that various types of bacteria may have played a role in the creation of gold deposits throughout the world.

TINY PROSPECTORS

Gold mines in Australia have added to the evidence for the historical role of bacteria in gold production. For his PhD, Reith studied communities of organisms existing in biofilms on the surface of gold grains from two mines. Biofilms from both mines contained *C. metallidurans* that thrived in the presence of toxic gold complexes. Later studies revealed that the biofilms on some of the grains also contained another bacterium, *Delftia acidovorans*, along with gold nanoparticles².

Tagging the bacterial DNA enabled the team to monitor the biofilms' chemical activity. "We saw the beautiful active biofilm dissolving the gold," says Reith's colleague Joël Brugger, a geochemist at the South Australian Museum in Adelaide. Some of the dissolved gold was redeposited into nanoparticles that were purer than the original grains. The nanoparticles can be transported through rocks and soils to end up in secondary deposits in cracks and crevices — much as Lengke found.

This dissolve-and-deposit process won't increase the total global supply of gold, but it does suggest two fresh approaches to discovering lodes of gold. "Bioindicator systems can be developed where the detection of certain microorganisms, microbial communities or functions indicates the presence of specific metals," Reith explains.

By using microarrays and DNA sequencing, for example, Reith and geochemist Carla Zammit of the University of Adelaide's Institute for Mineral and Energy Resources, along with fellow researchers, recently showed a correlation between the composition of microorganisms at mining sites and the metals present. The techniques they used included transcriptomic



In *The Great Work of the Metal Lover*, bacteria produce 24-carat gold for an audience.

microarrays, to detect up-regulation of gold-inducible genes of bacteria naturally associated with metals; this can indicate what metals are present. The team also used proteomic methods coupled with mass spectrometry to detect gold-binding proteins. Metabolomics enabled them to study the metabolites from biochemical reactions inside bacteria to access the 'chemical fingerprint' of cellular reactions involving metals. "As the costs of these techniques decrease in the future," Reith says, "exploration teams will employ molecular techniques to determine the microbial composition of a site and use this to identify possible sites of mineralization" — places where gold might be found.

Reith's group has shown that genetic information can distinguish between mineralized and unmineralized soils. "There are a few false positives," Brugger notes. "But in general the separation based on genetic information is quite encouraging."

Studies of the link between bacteria and gold are stimulating the development of hand-held biosensors. These will be able to analyse environmental samples for the presence of gold using gold-binding proteins derived from *C. metallidurans* and other bacteria known to coexist with gold — and hence find the precious metal. "Tests show that we reach detection limits of parts per billion in lab solutions with our whole-cell sensor," Reith says. "Exploration teams will be able to obtain information on gold concentrations in an environmental sample on-site, rather than after weeks of laboratory analysis." Reith's team is also developing molecular techniques, such as specific transcriptomic microarrays, that will be able to identify genes from *C. metallidurans* for use in a gold biosensor.

A RICHER HARVEST

Bacteria could also improve the output of established gold mines that have become uneconomic because they contain low-grade ore. Traditional technology leaves behind up to 20% of the available gold (see 'Extreme prospects', page S4). But it might be possible to extract some of that metal by putting bacteria to work. "Using microbial incubation of waste rock piles containing this unrecoverable gold might be a good way to achieve a low-tech, cost-efficient approach to concentrate gold into more recoverable forms," Reith says.

Reith and co-workers from the universities of Adelaide and Halle, Germany, for example, have started studying the biochemical process that catalyses the precipitation of gold from solution in *C. metallidurans*. The research involves the use of a synchrotron and transmission electron microscopy coupled with microarrays to detect the proteins that catalyse the reactions. Reith's team has also developed a method based on high-performance liquid chromatography and inductively coupled mass spectrometry to measure the complexed forms — such as gold chloride or gold thiosulphate — in which gold exists in aqueous solutions³. "This is important," Reith says, "because the type of ligand the gold is complexed with determines the mobility, as well as the environmental toxicity, of the complexes, which in turn is important for the biochemical reaction of the bacteria." The goal is to understand how gold exists in groundwater systems.

Researchers from McMaster University in Hamilton, Ontario, and Western Ontario University in London, Canada, recently discovered how *Delftia acidovorans* protects itself from soluble gold compounds. It does this by generating solid gold. The finding, the team notes, is the first demonstration that a secreted metabolite can protect against toxic gold and cause gold biomineralization⁴.

Lengke hopes bacteria can be coaxed to produce more of the metal. "Research should focus on better understanding of the mechanisms of gold precipitation, which will enable us to achieve better control over size, shape and uniformity," she says. "That will lead to the development of production-level amounts of gold."

That's a prospect for far in the future, but it would provide a suitable conclusion to a long-running process. "Microbes have been perfecting their biochemical pathways for more than 3.5 billion years," says Zammit. "And we're just starting to realize their potential." ■

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