

Deep Dwellers

Microbes thrive far below ground

By RICHARD MONASTERSKY

Tullis C. Onstott got a close-up peek at hell last year when he descended into the steamy bowels of the planet. With his shirt drenched from the 100 percent humidity, the Princeton University geologist slogged through the deepest gold mine in South Africa, where the temperature of the rock reaches 60°C (140°F) and sunlight is but a distant dream, some 3.5 kilometers up. After an hour of hiking through the passages, Onstott reached a recently blasted section of tunnel and took a hammer to the wall, knocking loose nuggets of rock that had spent the last 3 billion years locked underground. These chunks, he suspected, were alive inside.

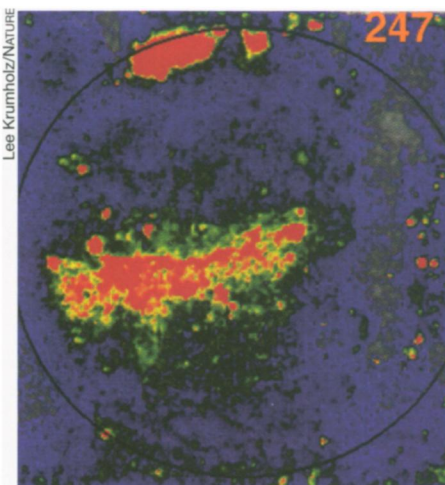
A little over 10 years ago, such an idea would have seemed pure folly. After all, everyone knew that life inhabits only a thin veneer of territory at Earth's surface. The oceans, the air, the ground, and even the soil teems with animals, plants, and microorganisms, but rock deep underground—with its scorching temperatures and toxic chemicals—is hardly a hospitable place for life.

Geologists and microbiologists have since buried that conventional wisdom. In the late 1980s, researchers sponsored by the Department of Energy found microbes living in rock 500 meters below the surface in South Carolina. In the last 4 years, Onstott and other researchers have pushed the envelope of life much deeper, extending it to nearly 3 km below ground. In some cases, these microbes have apparently remained prisoners of the deep for millions of years, making such colonies veritable living fossils.

"This is a revolution that's expanding the limits under which we know life to exist," says Onstott. "I think we're recognizing that life is a bit more tenacious than we had given it credit for, and there's a much broader realm of possibilities where life can survive."

The discoveries not only redefine how scientists view the modern Earth, they also raise intriguing questions about the origin of life and the possibility that microbes survive today beneath the surface of Mars and other planets.

Deep biology—the study of subsurface bacteria and similar organisms called archaea—has its roots in the 1920s, when a geologist and a microbiologist from the University of Chicago collected bacteria from oil deposits in sedimentary rocks 600 m below ground. The two scientists posited that these subterranean organisms could have evolved from microbes that were



Residents of sediments: Active bacteria (red and yellow) live in rock drilled in New Mexico.

buried 340 million years ago, when the sedimentary rocks initially formed.

Their ideas met only criticism. It was far more likely, other scientists argued, that the microbes were ordinary surface dwellers that had been transported into the sediments by the oil-drilling process, much as a dirty needle might inject a virus into a healthy person.

Support for the concept of deep biology resurfaced several times in the middle of the century, as scientists studied microbes from sediments beneath the seafloor. Yet the same concern about contamination quickly sank this research.

Oddly, it was contamination of a different sort that sparked the recent discoveries of subterranean life. In the 1980s, DOE came under pressure to address its toxic waste problems at various nuclear facilities. Uranium, heavy metals, and organic compounds were contaminating

groundwater deposits at these sites because the department had improperly discarded such materials during the Cold War drive to produce plutonium for bombs.

Deep microbes, if they existed, could affect the waste problem by either hindering or aiding the spread of toxic materials through the ground. So in 1985, Frank J. Wobber of DOE launched a subsurface science program, part of which aimed to assess the existence of life in rock. The researchers developed special anticontamination procedures for drilling into the ground and obtaining rock samples.

The DOE-sponsored team tested these techniques by drilling several boreholes at the Savannah River nuclear processing facility in South Carolina. By 1989, Wobber, Tommy J. Phelps of the Oak Ridge (Tenn.) National Laboratory, and their colleagues documented the existence of a diverse community of bacteria and archaea at a depth of 500 m.

The Savannah River discoveries seemed remarkable until Phelps, Onstott, and others studied rocks from the Taylorsville Basin in eastern Virginia. Joining forces with Texaco Oil Co., which was drilling exploratory wells in the basin, the scientists tested for microbial residents in much deeper rocks.

In 1993, the team collected new species of bacteria from 2.7 km below the ground—the current well-documented record for subsurface life. The scientists discussed the Taylorsville project in San Francisco last December at a meeting of the American Geophysical Union.

The organisms living beneath the Virginia landscape bear little resemblance to the typical microbes inhabiting the surface of the planet. The deep bacteria are known as thermophiles, or heat lovers, because their home has an ambient temperature of 75°C (167°F). One of the newly identified bacteria has earned the name *Bacillus infernus*, for its hellish habitat. Similar thermophilic microbes colonize volcanically heated springs, such as those in Yellowstone National Park, and scalding geysers on the ocean floor.

Onstott lists several lines of evidence, apart from their thermophilic nature, indicating that the Taylorsville microbes are true dwellers of the deep and not just contaminants from the surface.

The biggest potential source of contamination, the scientists reasoned, should be the lubricating fluids that drillers pump into the hole. Indeed, tests on these fluids revealed an abundance of aerobic microbes, which require oxygen to survive. Bacteria cultured from the deep rock samples were a different sort entirely: They were anaerobic organisms, which die when exposed to oxygen. This is exactly the type of bugs one would expect

to find in the oxygenless depths of the crust, says Onstott.

In another test, the scientists added various tracers to the drilling fluids. By checking rock samples for these human-made chemicals, they could identify which pieces had been contaminated by the drilling fluids.

Finally, the researchers measured the size of pores between mineral grains in the rock samples. Contaminated samples had relatively large pores that allowed drilling fluid to enter the rock. In the uncontaminated pieces, the conduits between pores were one-tenth the size of the bacteria, which would have barred any foreign microbes from penetrating the rock.

If bacteria couldn't get into the rock, they also couldn't leave. The tightly spaced mineral grains have served as cages, trapping any indigenous bacteria within the rock for millions of years, says Onstott.

The organisms survive by living on a spare diet of petroleum and other organic compounds dissolved in the groundwater. Because these nutrients are so dilute, the microbial colonies do not receive enough food to grow or reproduce, nor do they have room to spread through the rock. They simply live in a sort of suspended animation, say the scientists.

"What they're doing down there is not much. They're just trying to hold on," says Phelps.

Another colleague, James K. Fredrickson of Battelle Pacific Northwest Laboratory in Richland, Wash., says that the deep colonies are forcing microbiologists to reconsider their definition of survival. "We think of things having to grow in order to survive. That may be a good paradigm for life on the surface, but it may not be a good paradigm for bacteria in some environments. In fact, they may be able to sustain long periods without having to divide," Fredrickson says.

Evidence suggests that the microbes have been trapped for 80 million years, and possibly as long as 160 million years, says Onstott. This means that these species of bacteria may have become locked within the rock while the thunderous footsteps of dinosaurs were resounding overhead.

The DOE program has since documented other deep-dwelling communities in a sandstone and shale formation in New Mexico. They describe these bacteria in the March 6 *NATURE*.

Although the Virginia and New Mexico microbes endure a spartan existence, their lives seem relatively easy. That's because these organisms inhabit sedimentary rocks, which contain organic compounds that provide energy-rich food for the microbes. Other deep communities have no such luck.

Scientists have recently discovered microorganisms living within igneous formations—extremely hard rock lacking organic nutrients. The industrious residents of these rocks fashion their own organic molecules out of the barest of inorganic materials.

Relying only on hydrogen, water, and carbon dioxide—all of which are products of Earth's interior—these microbes are unique among the vast array of living species. All other organisms depend to some extent on the sun's energy, which is harnessed through photosynthesis and creates food for surface life. Even organisms living near seafloor vents rely indirectly on the oxygen supplied by photosynthesis, says Todd O. Stevens of Battelle.

Stevens and his colleague James P. McKinley discovered the first examples of microbial communities sustained by igneous rocks—known as lithoautotrophs—while studying basaltic lava flows near the Hanford nuclear processing facility in 1995 (SN: 10/21/95, p. 263). Since then, Swedish researchers have documented a similar community of lithoautotrophic bacteria and archaea living 400 m deep in a granite formation in southeast Sweden. They presented their findings last September at a meeting in Switzerland.

The new finds are significant because granite is one of the most abundant rocks on the continents, suggesting that these organisms are quite widespread, says Karsten Pedersen of Göteborg University in Sweden.

Although knowledge about the subsurface biosphere is just now starting to bloom, rough calculations suggest that Earth's upper crust is rife with life. Bacteria and archaea may reach as far as 4 km below the continental crust and 7 km into the oceanic crust, says Fredrickson. Deeper still, the rock is presumably too hot for life, which is not known to survive temperatures above 113°C.

Adding it all together, there could be as much life hidden below ground as there is above. Thomas Gold of Cornell University, a steadfast proponent of deep life, has calculated that the weight of all subterranean microbes could equal that of all organisms above the surface.

Biologists say that their deep discoveries have the broadest possible implications, stretching backward in time and outward into the distant reaches of space. If bacteria and archaea can survive today far below ground without any help from the surface, then it's possible that life could have started there, says Pedersen. He notes that the surface was a particularly nasty place early in Earth's history, subject to repeated assaults by giant meteorites and a high dose of ultraviolet rays from the sun. Deep rocks would have provided protection from the sterilizing temperatures and radiation.

"In the beginning of life on Earth, there



Some like it hot: Cells of *Bacillus infernus*.

was an enormous meteorite bombardment, which could have dried out the seas. That would have been a problem [for surface life], but if you go down 500 meters below ground, it would have been quite nice," says Pedersen.

Even now, life may be hiding beneath the dusty red surface of Mars and bodies even more distant. "If primary production can occur underground, where it is disconnected from photosynthesis, then there is no reason that [subsurface] life couldn't exist on several planets in the solar system," says Stevens. The surface of Mars is inhospitable because it lacks liquid water, but fluids may course through the warmer interior of the planet.

Deep life could also hold practical significance by helping solve toxic waste problems. The Swedish government, for instance, funds research on microbes to assess how they will influence an underground repository for nuclear waste. Pedersen's work suggests that the microbes alter groundwater chemistry in ways that would inhibit corrosion of containment canisters.

Subsurface microbes could also provide industry with novel routes for cleaning wastewater, says Onstott. Many of these microbes consume otherwise toxic chemicals and would thrive in industrial wastewaters, which are often quite hot and have high concentrations of dissolved salts.

Last year, DOE abruptly ended its research on subsurface microbes, even as the pace of discoveries was picking up. Researchers in the United States are now seeking support from other federal agencies to continue their quest for subsurface life on the continents, under the seafloor, and eventually on other planets.

Basic questions and practical concerns are driving investigators to explore new environments, such as the South African gold mine that Onstott braved last year. Preliminary tests on rocks from the mine reveal that thermophilic bacteria somehow manage to survive, even at the extreme depths of 3.5 km.

The geologist and his colleagues plan to return there soon, hoping to get to the bottom of Earth's most distant life. □