

Got a Toxic Mess? Call in the Microbes

Chlorinated solvents are nasty chemicals. But in the 1960s and 1970s, before anyone realized just how nasty they were, the United States government and some industrial manufacturers used the solvents freely, often dumping the leftovers in landfills or “out the back door.”

Today, the solvents are some of the most prevalent contaminants in groundwater in the United States. The chemicals are thought to cause cancer, and they’re expensive to clean up.

But oddly enough, some bacteria are already cleaning them up. These microbes live in soil and groundwater, and they “eat” the solvents, turning them into harmless substances. The federal government and some private companies are using the bacteria, called *Dehalococcoides*, to clean up toxic messes by putting them en masse into groundwater at contaminated sites.

A commercial test is available that can detect whether the helpful bacteria are present in soil. If there aren’t enough there, a company, SiREM, can provide billions of the bacteria that are ready to be transferred into the contaminated site.



At a site in Indiana, a worker injects the bacteria from steel containers into a pipe that goes underground. The smaller vessel is full of gas that helps push the bacteria down.

Image courtesy SiREM.

A Eureka Moment

Twenty years ago, General Electric (GE), Dupont, and other manufacturing companies began looking for inexpensive “natural” ways to clean up chlorinated solvents that they had dumped in their own back yards.

Around the same time, Cornell University engineer James Gossett, who had been working with the Air Force at its toxic waste sites, made a breakthrough. He found “something” in a sample from a sewage treatment plant in Ithaca, New York, that cleaned up chlorinated solvents. At the time, these chemicals were still sometimes dumped down the drain. Years later, in 1997, Gossett and Cornell biologist Stephen Zinder isolated the bacterium from its environment, and they published a paper describing the organism Zinder later named *Dehalococcoides ethenogenes*.

When scientists at Dupont and GE saw the paper, they could not believe their eyes. The Cornell researchers had sequenced the DNA of the same organism that they had found in their samples at their toxic sites. It was a Eureka moment for everyone. Not only could *Dehalococcoides* clean up chlorinated solvents, but also the bacteria live almost anywhere these chemicals are dumped. “When we discovered the bacterium, it made quite a splash,” says Zinder. “No one knew what to make of it,” he adds. “The bacterium looked promising, but was it applicable to the real world?”



The bacteria come in stainless steel vessels that contain roughly 2000 billion *Dehalococcoides* bacteria ready for injection into groundwater. Image courtesy SiREM.

Before adding the bacteria, the researchers infused the groundwater with ethanol to create the ideal environment for them. A few months later, they saw marked improvements in the level of ethene, an indicator that the bacteria were breaking down the solvents.

There was so much improvement that the EPA allowed the engineers to turn off one of the wells connected to the pump-and-treat system. Eventually, the engineers hope to shut down the system down completely.

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Bacteria Underfoot

Today, in the United States, the bacteria have been used to clean up chlorinated solvents in ten states at 17 sites including Kelly Air Force Base in Texas and Caldwell Trucking Superfund Site in New Jersey. For SiREM, business has been good of late; 13 of the sites were done in the past 18 months.

Chris Voci, a hydrogeologist at O’Brien and Gere Engineers in Blue Bell, Pennsylvania, uses the bacteria at a former electronics manufacturing company in Western

Pennsylvania whose cleanup of chlorinated solvents is regulated by the Environmental Protection Agency and managed by O'Brien and Gere.

A "pump-and-treat" system was originally installed to remove contaminants from the groundwater, but it would take decades to complete the task. So, Voci and his colleagues decided to enlist the help of SiREM's *Dehalococcoides* bacteria. Before adding the bacteria, the researchers infused the groundwater with ethanol to create the ideal environment for them. A few months later, they saw marked improvements in the level of ethene, an indicator that the bacteria were breaking down the solvents.

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Everyone Gets On Board

Both the public and private sectors have vested interests in making this microbe work. The number of contaminated sites in the United States is split right down the middle between the U.S. military and private companies. Various companies, universities, the EPA, the Department of Defense, and the Department of Energy have formed a consortium to develop real solutions to cleaning up chlorinated solvents. Once *Dehalococcoides* was discovered, the consortium began a pilot project to study the activity of the bacteria at Dover Air Force Base, where chlorinated solvents had been dumped. Researchers for the consortium successfully cleaned up the chemicals in a test project at Dover by adding *Dehalococcoides* bacteria to the groundwater. Federal regulators seemed pleased with the progress. In 2000, the EPA issued a memorandum

that outlined how this type of clean up, called bioaugmentation, is allowed under the EPA's Resource Conservation and Recovery Act. Individual states regulate the use of these bacteria at most sites.

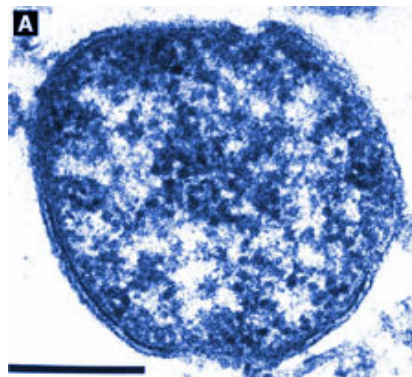
Meanwhile, at Dupont, scientists began developing a test to detect whether the bacteria were present in soil or groundwater. If there are enough bacteria, then they should clean up the chemicals on their own.

Dupont wanted to use the test itself, but the company also wanted a return on its investment. Company scientist Edwin Hendrickson created a test to detect DNA sequences specific to *Dehalococcoides*. Dupont eventually patented the test and now licenses the patent to SiREM in Canada, which markets the test worldwide.

Exploring the Genome

“The test is only the beginning,” says Hendrickson. “Right now, we can show that the organism is there, but we know little about what it does when we put it in the ground.”

To answer these questions, researchers at The Institute for Genomic Research (TIGR) in Rockville, Maryland, are sequencing the genome of *Dehalococcoides ethenogenes* with funding from the U.S. Department of Energy.



Dehalococcoides is round, flat and about 5 microns long. A micron is one thousandth of a millimeter.

Image courtesy *Science*.

To answer these questions, researchers at The Institute for Genomic Research (TIGR) in Rockville, Maryland, are sequencing the genome of *Dehalococcoides ethenogenes* with funding from the U.S. Department of Energy. By analyzing the bacterium's genome, the team, led by Rekha Seshadri at TIGR, has identified some 15 genes involved in breaking down chlorinated solvents. They are searching for others that will reveal how the bacterium grows—and also how it “eats” these chemicals underground. Still, some pretty basic questions remain: Why does *Dehalococcoides ethenogenes* do this? The bacterium has a small genome and can't do much more than break down chlorinated solvents. What did it eat before people began dumping their toxic messes in landfills? “It seems that the bacterium was born to dechlorinate,” says Zinder, who is collaborating with TIGR on the project.

Fears about “Franken-bugs”

David Ellis, who runs Dupont's program to develop new ways to clean up toxic chemicals, says that he doesn't think the general public is aware of this bacterium and its potential to break down chlorinated solvents.

Do some people have concerns? “Yes,” says Ellis. “There are sometimes concerns about whether these bugs are going to crawl out of the ground and eat my pipes and my kids, but luckily there are a lot of good answers to these fears.”

Most of the people making decisions about the use of these microbes are scientists who are educated about the “Franken-bug” fears, notes John Wilson of the EPA's National Risk Management Laboratory in Cincinnati, Ohio. The microbes are natural and have not been genetically modified in any way.

“We usually don't have to assuage someone's fears about the technology,” Wilson adds.

New technologies always raise questions. No one knows for sure what happens to the bacteria once all the toxic chemicals are cleaned up at a site, and no studies have been conducted. Do they die? Migrate to other sites? Or do they just stay in the ground?

Down the Road

Dehalococcoides won't solve many of the toxic waste problems. The bacteria are effective only on chlorinated solvents, and not other chemicals. In addition, many toxic sites are already fitted with expensive pump-and-treat technologies and their owners may not want to abandon them for other remedies.

"The bacterium is just one tool in the toolbox," says Wilson.

Even so, it is one of the few microbes being used extensively by the public and private sectors to clean up toxic chemicals.

"The U.S. government has more problems [with chlorinated solvents] than anyone else in the country," says Ellis. "The U.S. taxpayers stand to be some of the major beneficiaries of this microbe."

References:

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