Chemistry & Biology

Nature Cure: Bioremediation As a Sustainable Solution for Polluted Sites

Chandra Shekhar DOI 10.1016/j.chembiol.2012.03.004

Polluting is usually cheap and easy; cleaning up is often expensive and difficult. The traditional way to deal with contaminated soil or sediment is to dig it out and dispose of it elsewhere. Contaminated water is usually pumped out, treated, and released back. Results are often unsatisfactory-studies show that only a small fraction of sites get acceptably cleaned up and the environment is rarely restored. Further, these measures are expensive: using them to restore all contaminated sites in the US could easily cost more than a trillion dollars. In contrast, bioremediation-treating contaminants using natural methods-can

short of the worst-case scenario. Here again we have to thank naturally occurring oil-hungry bugs with a taste for petroleum acquired from the many natural oil seeps in the Gulf for helping to mitigate another major oil catastrophe. "Within three weeks of capping the oil, there was basically no slick anymore," says Larry Wackett, Ph.D., of the University of Minnesota. "It was pretty amazing how quickly that happened."

Most pollutants released by human activity already occur in nature, so it comes as no surprise that microbes can metabolize them. Many bacteria consume polycyclic aromatic hydrocarbons (PAHs),

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potentially restore the ecology of polluted sites at a fraction of this cost. "Unlike a brute force approach, bioremediation can help make a very specific change to the system so that it heals," says Frank Loeffler, Ph.D., of the University of Tennessee. "It has the potential to be not only cheaper but also more environment friendly and hopefully more acceptable to regulators and the public."

Two major oil spills in recent years illustrate this potential. In 1989, the tanker Exxon Valdez ran aground off the Alaskan coast, spilling more than ten million gallons of crude oil. Now, more than two decades later, most of the spilled oil has vanished, thanks largely to natural microbial processes. In the 2010 Deepwater Horizon accident, an exploratory oil well in the Gulf of Mexico about 50 miles off the US coast blew open and spewed out an estimated 200 million gallons of crude. An ecological calamity of nightmarish proportions was predicted. However, while the environmental impact has indeed been extremely serious, it has fallen well

which are carcinogenic compounds released by partial combustion of many fuels, and release harmless byproducts. Other bacteria and fungi can digest explosives such as TNT and render them innocuous. Chlorinated solvents such as perchloroethene (PCE) and trichloroethene (TCE) are another major pollutant class. Stable, poorly soluble in water, and rare in nature, such compounds were believed to be microbe proof. In the late 1980s, however, evidence emerged of rare, unusual microbes that could pull the chlorine out of PCE and turn it into ethene, a harmless hydrocarbon gas. These are now classified as "organohalide respirers," bacteria that eat hydrogen and breathe chlorinated compounds instead of oxygen. Since their discovery, these unique bugs have been used by environmental engineering companies to clean up hundreds of sites contaminated with chlorinated solvents. "This is a good example of how the discovery of a single new organism could change engineering within a few years," says Loeffler.

For contaminant-degrading microbes to do their job, nutrients such as oxygen, carbon, nitrogen, and phosphorus should be present in the right concentrations. If not, adding chemical supplements-"biostimulation" -could help. In the aftermath of the Exxon Valdez spill, field tests indicated that increasing nitrogen levels would boost the rate of oil degradation. Subsequently, nearly 50,000 kg of nitrogenous fertilizer was applied to more than 2,000 separate shoreline locations over a 3 year period, in perhaps the biggest bioremediation effort in history. This is believed to have been a major factor in the eventual disappearance of the oil. Soap-like compounds (surfactants) and dispersants such as the ones used in the Deepwater Horizon spill could also help microbes by breaking up, loosening, or diluting contaminants. "We establish a partnership with the microorganisms," says Bruce Rittmann, Ph.D., of the Arizona State University in Tempe, who has invented a water purification system based on this concept. In his setup, hydrogen gas is delivered directly to bacteria that then breathe-and thereby break down-TCE, nitrates, and similar substances in the water. "We create a happy home life for the microbes, and in return they get rid of the contaminants for us."

Of course, pampering microbes will pay off only when they are capable of destroying the target contaminant. In the absence of such bugs, one could try "bioaugmenting" the site with foreign microbial species that have the requisite capability. In theory, the introduced bugs would not only attack the pollutant but also transmit the relevant skills to native species. Except in the case of chlorinated solvents, however, bioaugmentation has enjoyed limited success in the field. Foreign bugs rarely survive long enough to do their job. Further, site owners and regulators often have concerns about releasing such organisms, especially genetically modified

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ones. One way to sidestep these concerns is to use enzymes instead of live microbes. Wackett, for example, uses silica beads to encapsulate recombinant bacteria that make large amounts of an enzyme that degrades the herbicide atrazine. Cooking the beads to 45°C kills the cells but not their atrazine-degrading capability, Wackett says. "We are essentially using the dead cell as a bag of enzymes."

Under microbial influence, many organic pollutants break down into water, carbon dioxide, and other harmless substances. In contrast, inorganic pollutants-heavy metals such as mercury and lead or radionuclides such as uranium and plutonium-don't degrade and can't be destroyed. The goal of remediation then is to convert such contaminants into relatively less harmful forms. Sulfides of cadmium or copper, for example, are less soluble and therefore less toxic than the corresponding sulfates. In 2009, researchers led by Geoffrey Gadd, Ph.D., of the University of Dundee in Scotland showed that several species of fungi could transform depleted uranium, a toxic alloy used in weaponry, into more stable uranyl phosphate minerals. "Before this finding you would never have associated fungi with making such minerals." says Gadd. However, he has also found that microbes can do the reverse, breaking down insoluble minerals into more soluble, toxic components; for example, a common soil fungus can corrode pyromorphite, a relatively safe lead mineral, and release harmful lead ions. "Nothing can be completely stable where microbes are concerned," Gadd says.

For inorganic contamination, remediation using plants, "phytoremediation," may be a better option. Plants could help simply by holding soil and sediments in place and drawing groundwater out, thus reducing contaminant escape. They can lock up toxic elements in their roots, extract and store them in their aboveground parts, or blow them off as vapor. Modified tobacco plants, for example, can absorb methyl mercury and vaporize it in the less harmful metallic form;

conversely, some mustard plants can take toxic metallic selenium and emit it in its safer methyl form. Some fern and mustard species can extract and store significant amounts (up to 1 g/kg) of arsenic, selenium, and other elements. The detoxifying capabilities of many plants get a boost from microorganisms that live on their root systems; for example, a root-colonizing fungus discovered in a copper mine appears to help pine trees remediate copper contamination. "The combined efforts of plants and microorganisms will be able to do wonders," says M.N.V. Prasad, Ph.D., of the University of Hyderabad in India.

Even with the help of microbial and plant allies, cleaning up contamination will remain a major challenge due to the sheer diversity of pollution scenarios. Each site has a unique geology, with its individual pattern of soil layers, sediment, and water flow. It will also have a unique distribution of temperature, permeability, texture, pH, oxygen level, chemical composition, microbial communities, and other factors. Each type of pollutant may need a different set of conditions to degrade. Further, locating the source of pollution at a site is often tricky. Heavierthan-water liquids such as chlorinated solvents may trickle through the surface layers and pool onto the clay or bedrock underneath. "It often goes so deep we can't dig it out and goes in all kinds of directions so that we can't even find it," says bioremediation pioneer Perry McCarty, Sc.D., of Stanford University. Digging holes to take samples is very expensive, so engineers are often forced to guess which way a plume of contamination is moving. Cleaning up a light, volatile substance such as gasoline from loose surface media such as sand or gravel is relatively easy. In contrast, once a dense, stable, water-repelling liquid such as PCE has seeped into clay or fractured rock below the water table, it becomes virtually impossible to dislodge. The compound might then foul up the groundwater for decades. "One of the biggest lessons we've learned over the past 20–30 years is how difficult and expensive it is to clean up ground water once it's contaminated," says McCarty. "So the best approach is prevention."

While strict environmental regulations may prevent future contamination, a vast number of sites worldwide remain significantly polluted from past human activities. A growing world population will increase the demand to reclaim these places. The market for innovative remediation methods is thus potentially huge but also highly uncertain. "Everybody expects clean air, water, and soil, but nobody wants to pay for their cleanup," says Loeffler. Despite this challenge, the bioremediation sector is growing. New knowledge is constantly emerging about microbial species, pathways, and enzymes relevant to bioremediation; a database encapsulating this is being maintained by Wackett's group at the University of Minnesota. Researchers are finding new ways to exploit microbial mechanisms such as chemotaxis, biofilm formation, and biosurfactant production. Gary Sayler, Ph.D., of the University of Tennessee has developed a recombinant bacterium that glows when it is degrading pollutants. "If the practicing engineer could keep the lights turned on, we can achieve maximum rates of degradation," he says. Robert Borden, Ph.D., of the North Carolina State University has pioneered the use of emulsified vegetable oil as a steady nutrient source for contaminant-degrading organisms. "We have seen a whole battery of innovations in recent years, such as improved biostimulants, environmental nanomaterials, and electrobioremediation" says J.-Julio Ortega-Calvo, Ph.D., member of the Spanish National Research Council and the European council of the Society of Environmental Toxicology and Chemistry (SETAC). "While some problems remain unsolved, bioremediation is today a safe, low-cost alternative for cleaning up polluted sites."

Chandra Shekhar (chandra@nasw.org) is a science writer based in Princeton NJ.