For three decades starting in the 1940s, General Electric dumped solvents from its manufacturing facilities into New York’s Hudson River, contaminating it with polychlorinated biphenyls (PCBs). Scientists worried about how best to clean up the pollutants. “At that time, they thought PCBs were completely nonbiodegradable,” says Lawrence P. Wackett, a biochemist at the University of Minnesota Twin Cities who consulted for the company in the late 1980s.

But analysis of sediment cores extracted from the river throughout the 1980s showed that the PCBs were slowly losing their chlorine atoms and turning into benign hydrocarbons. Later, scientists determined that the transformation was performed by microbes.

Now, researchers are hoping microbes could do the same for per- and polyfluoroalkyl substances (PFAS), also known as forever chemicals. Used in personal care products as well as firefighting foam, stain-repellent coatings, and membranes for chlor-alkali production, PFAS have strong carbon–fluorine bonds that make them difficult to degrade. PFAS have become a high-profile contaminant, polluting areas near manufacturing facilities that make or use them and military sites like air bases. Researchers are still trying to fully understand the health effects of PFAS but have determined that some are carcinogenic and toxic to multiple systems.

Traditionally, to treat contaminated water, remediers first concentrate the PFAS, typically via an activated carbon filter, and then incinerate the saturated filters at high temperatures. Biological remediation could be more cost-effective for low levels of PFAS in large volumes of contaminated material. So far, no organism has been found that can completely defluorinate PFAS, but researchers have no reason to believe that microbes couldn’t eventually do the job. “Never say never in terms of what they can handle,” Wackett says.

The US Department of Defense’s Strategic Environmental Research and Development Program (SERDP) has funded extensive research into the chemical and physical remediation of PFAS. In 2018, SERDP launched a call for research...
proposals exploring the biodegradation of PFAS and awarded $2.75 million to five projects beginning last year.

However, other scientists, like Rolf U. Halden, an environmental engineer at Arizona State University, worry that investigating bioremediation of PFAS distracts from the real work of having to dial back on their use. Halden is “very, very skeptical” that a microbe could be practically deployed to remediate PFAS. “I think that this will only delay the actual hard question that we have to answer: How much [PFAS] is too much, and how do we get to a healthy use of these very useful materials?” he says.

Microbial infallibility
In his 1951 textbook, The Chemical Activities of Bacteria, British microbiologist Ernest Gale put forward the microbial infallibility hypothesis—that if there is energy to be gained from a compound, a microorganism will figure out how to extract it and create a niche for itself. After the Deepwater Horizon explosion and oil spill, for example, scientists found that microbes eventually ate most of the energy-rich hydrocarbon compounds that spilled into the Gulf of Mexico, resolving part of the problem naturally.

Halogenated compounds such as PCBs contain much less energy than hydrocarbons, but microbes can still use them. In the Hudson River, Dehalococcoides bacteria living in oxygen-poor environments in the sediment transfer electrons to the PCBs, reducing them and kicking out chloride ions, Wackett explains.

In fact, Dehalococcoides are obligate dehalogenators, Halden says, meaning they can survive only by dumping their electrons on halogenated organic compounds. But there are significant differences between PFAS and their chlorinated counterparts, he says. To start, carbon–fluorine bonds are much stronger and harder to break than carbon–chlorine bonds.

More importantly, microbes evolved along with thousands of naturally occurring chlorinated compounds, so when bacteria like Dehalococcoides encounter human-made chlorinated pollutants like PCBs or trichloroethylene (TCE), they do not see those pollutants as completely foreign. “These organisms existed before we created TCE,” Halden says. But naturally occurring fluorinated compounds are rare; only fluoroacetate is well studied in plants, and it contains just one fluorine atom. PFAS, in contrast, especially the perfluorinated ones, are swathed in fluorine atoms. “That renders the chemicals almost bulletproof,” Halden says.

PFAS are so recalcitrant that one of the five SERDP-funded projects investigating biological methods to destroy PFAS instead demonstrated that previously reported chemical degradation methods don’t work. Pedro J. J. Alvarez, an environmental engineer at Rice University, had been working with bacteria that can produce copious amounts of superoxide outside their cells. Alvarez read that superoxide generated by decomposing hydrogen peroxide could break down perfluorooctanoic acid (PFOA), one of the most common PFAS found in the environment, so he proposed that superoxide-generating bacteria could perhaps degrade PFAS. He and his colleagues found, however, that superoxide generated chemically or enzymatically could not break down PFOA. When Alvarez and his team dug more deeply, they found that another heavily investigated substance for PFOA degradation, hydroxyl radicals, could not do the job either.

But Alvarez is not discouraged. “If it’s going to fail, let’s fail fast so that we do not waste time on this,” he says. That way, the research community can move on and try other approaches. His team’s recent work on using the superoxide-generating bacteria to dechlorinate TCE has shown outstanding results, Alvarez says. The bacteria are effective even when oxygen, which could compete with TCE as an electron acceptor, is present. Because TCE is a common co-contaminant with PFAS, Alvarez thinks the superoxide-generating bacteria could still prove useful by eliminating other pollutants that may interfere with remediation processes that target PFAS.

And Alvarez believes in the microbial infallibility hypothesis—that a microbe will find a way to use even tough compounds like PFAS. “I am certain that it can happen,” he says.

A point of weakness
A key factor for how easily a microbe can break down a fluorinated compound is if the molecule contains a spot vulnerable to attack, such as a carbon–hydrogen bond, says Jinxia Liu, an environmental engineer at McGill University. Liu has been investigating the biotransformation of polyfluorinated compounds called fluorotelomers for over a decade. Fluorotelomers such as 6:2 fluorotelomer sulfonate, which is used in firefighting foams, contain such a spot that is susceptible to microbial action.

Aerobic Gordonia bacteria perform a well-known transformation on fluorotelomers: they consume the sulfonated part, leaving a highly persistent, perfluorinated carboxylic acid. Liu has observed perfluorinated biotransformation products that are one or two carbons shorter, though, suggesting that Gordonia is also capable of chopping up the fluorinated tail one carbon at a time. “In theory, we can completely defluorinate a fluorotelomer,” says Liu, whose calculations show that the process is energetically favorable. But Liu’s team observed that the removal usually stops after two cycles. The researchers are working to figure out why
the defluorination stops and how to push the microbes to repeat the removal until all the fluorine atoms are gone.

Gordonia bacteria can chop up the long tails of fluorotelomers, such as 6:2 fluorotelomer sulfonyl acid (top) and 6:2 fluorotelomer sulfonamide alkylbetaine (bottom), one carbon at a time. Credit: C&EN.

In perfluorinated molecules, moieties like a double bond could serve as the necessary point of weakness. Yujie Men of the University of California, Riverside, incubated KB1, a commercially available microbial culture that is used for dechlorination and that includes Dehalococcoides bacteria, with lactate and a variety of perfluorinated molecules. The lactate provides electrons for the microbes, while the PFAS act as electron acceptors. After 180 days, 90% of the unsaturated perfluorinated molecules were degraded compared with none of the saturated perfluorinated ones.

A commercially available microbial culture could degrade unsaturated PFAS such as (E)-perfluoro(4-methylpent-2-enoic acid) (left) and 4,5,5,5-tetrafluoro-4-(trifluoromethyl)-2-pentenoic acid (right). Credit: C&EN.

Men’s team has identified intermediates suggesting that two initial reactions—with opposite effects—compete during the biotransformation process. The microbes could be replacing fluorine atoms on the double-bonded carbons with hydrogen, thereby making the molecule more vulnerable to additional defluorination. Or they could be adding two hydrogen atoms across the double bond, creating a saturated compound that is more resistant to defluorination. Men is working to identify the specific bacteria responsible for the reactions and the enzymes involved. “There is a way to direct them as long as we know which microorganism and which enzymes are carrying out the defluorination reaction and the hydrogenation reaction,” she says.

As for PFOA and perfluorooctanesulfonic acid (PFOS), which contain no weak spots, a bacterium native to the wetlands of New Jersey may be able to defluorinate them. Peter R. Jaffé and his group at Princeton University have been studying A6, a strain of the microbe Acidimicrobium, since 2005. This microbe performs a reaction called Feammox, in which it transfers electrons from ammonium ions to iron (III) ions in acidic soil.

Jaffé’s team sequenced A6’s genome and noticed it had genes coding for dehalogenases. “Some of them were quite novel,” Jaffé says. The team then decided to see what happens when A6 is given only PFAS as its sole source of carbon. Over 100 days of incubation with either PFOA or PFOS, the researchers found a steady disappearance of up to 60% of the compounds, with an accompanying rise in dissolved organic carbon and fluoride ions.

Jaffé is now collaborating with the University of Minnesota’s Wackett to decipher the mechanisms behind A6’s defluorinating power. Wackett is using a combination of computational and experimental techniques to narrow down the enzymes and the genes that could be responsible.

Practicality

In the laboratory, researchers can create ideal conditions for microbes to feed on PFAS or even force them to do it. Deploying microbes out in the environment for in situ remediation, however, presents significant challenges. Halden notes that Jaffé’s and Men’s studies used high concentrations of PFAS. But in the real-world environment, even though PFAS are present in some places at levels dangerous to people’s health, they still exist only at parts-per-billion concentrations.

Worse, there are many other goodies in the environment for microbes to feast on, and it is hard to control what they choose. Men has witnessed microbes in the commercial KB1 culture turn from consuming PFAS to eating an alternative like TCE when it is present. “It’s very difficult because it’s like you’re asking someone to eat grass rather than normal food,” Men says.

Whether microbial action can be exploited cost-effectively will also depend on the contaminated site, Alvarez says. A key parameter is how quickly water will carry PFAS at a particular site to another place: if the migration is slow,
remediators can get away with a slow microbial degradation process. “But if the rate of migration is fast, then you better look for a more aggressive, faster solution to protect public health,” he says.

A more feasible option is ex situ remediation, in which the contaminated material is pumped away and treated in an isolated bioreactor under controllable conditions. In a treatment facility, biological remediation can also be paired with chemical remediation. Bruce E. Rittmann, an environmental engineer at Arizona State University, is taking such a two-step approach.

“Our strategy is based on the understanding that we aren’t going to be able to directly biodegrade these PFAS,” he says. “We need to start the job for the microorganisms.”

In Rittmann’s strategy, PFAS first go through a hydrogenation reaction with a palladium catalyst, which replaces some of the fluorine atoms with hydrogen. Then, the partially defluorinated material is fed to a diverse group of microbes that finish the defluorination job. Rittmann has successfully demonstrated the steps individually and is now working to link them in a two-stage setup. The group is also investigating various research questions, such as how much defluorination is required in the first step and what the biotransformation products are.

Bruce Rittmann is taking a two-step approach to PFAS degradation, starting with a palladium catalyst to partially defluorinate (left tube) the PFAS followed by microbes to finish the job (right tube). Credit: Bruce Rittmann.

McGill’s Liu is collaborating with microbiologist Nancy N. Perreault of Canada’s National Research Council to explore microbial biodegradation of PFOS after it first receives photochemical treatment that partially defluorinates it.

Currently, the microbes being studied for PFAS degradation grow too slowly, and their defluorination performances are not good enough for any practical remedy for environmental contamination of PFAS, Men says. Nevertheless, at the risk of sounding too optimistic, Men says she is hopeful that microbes will eventually rise to the challenge: “Bacteria have really huge potential, and they evolve very fast.”

Whether or not microbes are ever able to conquer PFAS, most researchers agree that the use of PFAS should be restricted without delay. Despite voluntary phase-outs and even international bans on a few PFAS, some 1,400 PFAS are still being used in about 200 applications, spanning almost every industry. Some applications, such as medical equipment and chlor-alkali membranes, might justify the use of PFAS. But PFAS may not be required for other products—like artificial turf, guitar strings, or children’s rain jackets. “We have to look deeper into what’s really necessary and what is not,” Liu says.