Perchlorate Biodegradation
for WATER Treatment

Biological reactors currently treat nearly 30 million liters of perchlorate-contaminated groundwater per day, and in situ treatment techniques are now showing success at the field scale.

PAUL B. HATZINGER
SHAW ENVIRONMENTAL, INC.

Biological approaches for treating perchlorate are proving to be highly effective and economically attractive. During the past several years, numerous laboratory and field pilot studies have evaluated different bioreactor designs and in situ techniques for cleaning perchlorate-contaminated groundwater and wastewater. On the basis of these evaluations, several full-scale biological treatment systems now operate in the United States. These systems remove perchlorate from waters with concentrations ranging from ~2 to >4000 mg/L to effluent levels <4 µg/L. This article summarizes the recent progress in perchlorate bioremediation.

Fame circa 1997

The first indication that perchlorate contamination in groundwater was a significant environmental issue came in early 1997 after the California Department of Health Services (DHS) developed an improved analytical technique that reduced the minimum reporting level (MRL) for the anion in water from ~400 to 4 µg/L. Subsequent drinking-water testing performed throughout California revealed contamination in several regions of the state, including Los Angeles, San Bernardino, Riverside, Orange, and Sacramento counties. As of December 2004, 361 out of ~6800 public drinking-water sources in California have tested positive for perchlorate (*1*). Numerous private wells are also tainted. Although California appears to have the most widespread contamination, 34 other states, including Nevada, Arizona, Texas, Utah, New Mexico, Maryland, and Massachusetts, have reported perchlorate levels in groundwater or drinking water (*2*). The lower Colorado River, which supplies drinking water to ~15 million people, also contains measurable levels of the pollutant at certain times of the year (~4–9 µg/L; *3*).

L. KEITH BAILEY

The majority of the anthropogenic perchlorate contamination in groundwater stems from historical disposal practices by the aerospace and ordnance industries, the military, and chemical manufacturers. In some areas, the production and use of highway safety flares, fireworks, and explosives have also been linked to groundwater pollution. Perchlorate salts have been used in the U.S. defense and space programs for several decades as primary oxidants in the solid propellants that power rocket motors, rocket boosters, and missiles. In the era before strict environmental regulations, solid perchlorate-containing fuels requiring disposal were often burned in open-burn and open-detonation areas, and aqueous processing waters or wastewaters were released to surface soils or discharged into lagoons or evaporation ponds. Perchlorate is also present in many types of munitions and ordnance, and it has been detected in low concentrations in groundwater beneath some military training ranges (4).

Perchlorate has also long been known to be present naturally in nitrate deposits in the Atacama Desert of northern Chile (5). Nitrogen fertilizers from these deposits, once widely used, are still imported into the United States. The U.S. Geological Survey recently reported the presence of perchlorate in several other minerals in the southwestern United States, at levels as high as 3700 mg/kg (6). This finding has generated new interest in natural perchlorate sources. It also may provide an explanation for the detection of perchlorate in groundwater over a wide area of western Texas where military sources are unlikely (7). A recent U.S. Food and Drug Administration study in which perchlorate was detected at low levels in nearly all of the milk (3.2–10.4 µg/L) and lettuce (1.0–71.0 µg/L) samples collected from 16 states may also indicate natural perchlorate is much more prevalent in the environment than previously thought (8).

The level of perchlorate that is safe for human consumption has been the subject of intense scientific debate. The U.S. EPA released a draft toxicological review on perchlorate in 2002 that proposed a reference dose (RfD) of 0.03 µg perchlorate/kg body weight/day (µg/kg/day), which equates to ~1 µg/L in drinking water (9). After reviewing the available body of toxicological data, a committee of the National Academy of Sciences (NAS) more recently suggested that an RfD of 0.7 µg/kg/day would protect human health (10). EPA subsequently revised its draft RfD to meet the NAS recommendation and issued an official RfD of 0.7 µg/kg/day in February 2005. Under the same assumptions as the original EPA assessment, this RfD corresponds to a drinking-water-equivalent level (DWEL) of 24.5 µg/L. However, some toxicologists believe that levels as high as 220 µg/L in drinking water are safe (11).

EPA must now decide whether to develop a federal drinking-water standard for perchlorate based on Safe Drinking Water Act criteria. If promulgated, such a standard is unlikely to be higher than the DWEL of 24.5 µg/L, but it may be appreciably lower than this value, given the potential contribution of perchlorate from nondrinking-water sources such as milk and produce.

Most detections in water to date are <20 µg/L, and many are <10 µg/L (12, 13). Thus, determining perchlorate risk and the subsequent drinking-water standard has important economic implications. A federal drinking-water standard for perchlorate is not likely for several years. However, a number of states have set their own advisory levels, including Nevada (18 µg/L); Arizona (14 µg/L); California (6 µg/L); New York (5 µg/L); Texas (4 µg/L); and New Mexico, Massachusetts, and Maryland (1 µg/L).

Treatment technologies

Perchlorate is highly soluble in water, nonadsorptive, nonvolatile, and chemically stable. As a groundwater contaminant, its properties most closely resemble nitrate. Therefore, many traditional water treatment approaches, such as carbon adsorption, chemical oxidation, and air-stripping, that are used for solvents and other organic pollutants are ineffective for removing perchlorate from groundwater or drinking water (14, 15). Currently, the two primary technologies are physical removal through the use of ion-exchange resins and biological treatment with reactor systems. In situ biological treatment, including the construction of engineered cutoff barriers and the development of technologies to treat perchlorate source areas, is also rapidly emerging (16, 17).

The choice of technologies at a given site depends on numerous variables, including perchlorate concentration and plume characteristics, groundwater geochemistry, the presence of co-contaminants and existing treatment systems for those contaminants, the remedial objectives, the end use of the treated water, and economics. Additional information on ion exchange is available in recent papers (18, 19). The remainder of this article will focus on perchlorate biodegradation and the ex situ and in situ applications of this process for remediation.

Biodegradation

Bacterial reduction of perchlorate was observed as early as the 1950s, but most of the research in this area has been conducted only during the past several years as perchlorate emerged as an environmental contaminant. A wide variety of perchlorate-degrading strains have been isolated, many of which belong to the novel *Dechloromonas*, *Azospira* (formerly *Dechlorosoma*), and *Dechlorospirillum* genera (20–23). The organisms characterized to date are primarily Gram-negative, facultative anaerobes.

All of these bacteria can reduce chlorate, although some organisms capable of chlorate reduction cannot metabolize perchlorate, and most are denitrifiers. Pure culture studies have shown that molybdenum is essential for perchlorate metabolism and that both oxygen and nitrate biochemically inhibit the process in some strains (23–25). Perchlorate-reducing bacteria appear to be nearly ubiquitous in natural environments such as soils, sediments, surface water, and groundwater aqui-

fers (*22*, *23*, *26–28*). Why the physiological trait of perchlorate reduction is so prevalent in nature is unclear. However, new data suggesting that perchlorate may occur in low concentrations over wide areas, possibly from natural sources, could help to explain this observation.

Perchlorate-reducing organisms couple the oxidization of an organic or inorganic electron donor to the reduction of perchlorate in a form of anaerobic respiration (*29*). Various organic electron donors, including ethanol, fatty acids (e.g., acetate, lactate, propionate, and citrate), vegetable oils, and molasses support this metabolism. In addition, autotrophic strains that couple the oxidation of hydrogen gas to perchlorate reduction have now been reported (*21*, *30*). Perchlorate reduction produces chloride and oxygen as degradation products by the following pathway (*29*):

$$ClO_4^- \longrightarrow ClO_3^- \longrightarrow ClO_2^- \longrightarrow Cl^- + O_2$$
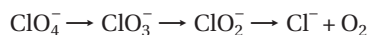
A two-enzyme system appears to catalyze the reaction. The initial enzyme, a reductase, reduces perchlorate to chlorate and then to chlorite (*31*). A second, highly conserved, chlorite dismutase then disproportionates the chlorite into chloride and oxygen, presumably as a detoxification reaction (*23*, *32*). The broad environmental distribution and diversity of perchlorate-reducing strains, combined with their ability to metabolize perchlorate to innocuous chloride and oxygen, have prompted rapid development and implementation of in situ and ex situ bioremediation strategies for perchlorate, as detailed in subsequent sections. (The phylogeny, ecology, and enzymology of perchlorate-reducing strains were recently reviewed [*23*, *30*].)

Treatment in bioreactors

Bioreactors are engineered systems that maintain high densities of pollutant-metabolizing organisms in contact with groundwater or wastewater. These systems can be divided into two main categories: suspended-growth reactors and fixed-film reactors, plus some hybrid designs (*33*). Suspended-growth systems, such as continuous-flow stirred-tank reactors (CSTRs) and most sequencing batch reactors, maintain organisms in suspension (Figure 1a). These systems have been used most frequently for the aerobic or anaerobic treatment of wastewaters, including chemical processing wastes and domestic municipal wastewater (i.e., the activated sludge process). This design has also been applied successfully at full scale for the treatment of perchlorate in concentrated wastewaters. Fixed-film systems, which include packed-bed reactors (PBRs), fluidized-bed reactors (FBRs), rotating biological contactors, and other designs, contain biofilms attached to solid media. These reactors are most commonly used to treat dilute contaminant streams and have been successfully implemented at pilot and full scales for remediating perchlorate-contaminated groundwater.

High-strength wastewater. The first bioreactor design tested for perchlorate treatment was a
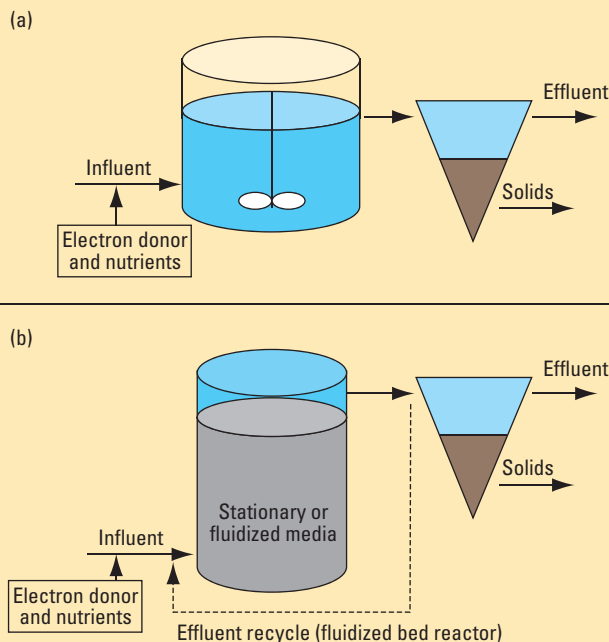
suspended-growth system. Developed in the early 1990s at the Air Force Research Laboratory, this reactor treats the wastewater generated when high-pressure water is used to remove solid fuels from rockets and missiles, a process often termed "hog out" (*34*). The laboratory system used a mixed microbial culture, from which the perchlorate-degrader *Wolinella succinogenes* HAP-1 was eventually isolated (*35*). The CSTR design was tested at pilot scale at Tyndall Air Force Base in Florida and then installed at the Thiokol rocket production facility in Brigham City, Utah, in 1997 (*36*).

The original Thiokol system consisted of two anaerobic CSTRs (6000 and 2700 L) and associated equipment for electron-donor feed and pH adjustment, process control, and effluent clarification and discharge. In 2002, two 3800-L reactors were added to increase capacity and permit the simultaneous treatment of 3 different effluent streams containing ammonium perchlorate, potassium perchlorate, and mixed nitrates, respectively. A cheese whey and yeast extract mixture was initially used as an electron donor, but it was subsequently replaced with desugared molasses to reduce cost and improve efficiency. The expanded system is capable of treating ~3600 kg perchlorate/month from influent concentrations ranging from 4000 to 5000 mg/L (achieved by diluting the concentrated wastewater) to effluent concentrations below the MRL for this matrix (~400 µg/L).

In 2003, the Hodgdon Powder Co. constructed a second suspended-growth reactor system at its gun-

powder manufacturing facility in Herington, Kan. This design, also consisting of the dual anaerobic CSTRs, is successfully treating gunpowder processing wastes containing perchlorate at influent levels >3000 mg/L to effluent levels that are below the MRL of ~20 µg/L. The two 9500-L molasses-fed reactors are designed to process 9500–19,000 L of wastewater daily. The operating and maintenance cost for this system depends on the actual perchlorate and nitrate composition of the feed water. However, for a nominal 1000-mg/L perchlorate wastewater, this cost (which includes nutrient, chemicals, power, and maintenance) is ~$10 per 3800 L.

The McGregor Naval Weapons Industrial Reserve Plant (McGregor, Texas).

In addition to being used for hog out and processing gunpowder wastewaters, a specialized suspended-growth reactor is currently under pilot evaluation in La Puente, Calif., for the treatment of ion-exchange brine, which is a nitrate- and perchlorate-laden salt brine produced during groundwater treatment by regenerable ion exchange (*19*). The system uses an internal membrane to separate biomass from effluent water and subsequently to maintain high cell densities within the reactor. During field-testing of the pilot system, reduction of perchlorate to below the applicable MRL of 40 µg/L was observed at salt concentrations >6%. If successful at full scale, this technology could substantially increase the application of regenerable ion exchange for perchlorate treatment. Given the absence of an effective and economically viable option for removing perchlorate from ion exchange brines, the regenerable systems have mostly been displaced by those that use disposable resins for perchlorate treatment.

Groundwater treatment. Two different fixed-film reactor designs have been tested for perchlorate treatment: PBRs and FBRs (Figure 1b). For biological growth, PBRs use stationary media, ranging from coarse sand to plastic sheet or ring packing. The medium is housed in a reactor vessel, and contaminated water is passed through it in an upflow or downflow manner. As in suspended-growth systems, an electron donor is mixed with the influent water to support microbial degradation of perchlorate, as well as of oxygen and nitrate. Microbial cells grow as a biofilm either attached to or within the packing material, and cells of the biofilm degrade perchlorate as groundwater passes through the reactor. The key advantage of these systems

over suspended-growth reactors is their ability to maintain high densities of biomass within the reactor, even in the presence of rapidly flowing groundwater or wastewater.

Several researchers have tested laboratory-scale PBRs for perchlorate treatment (*37–39*), and a few pilot-scale studies have also now been completed (*30, 40*). The most extensive pilot-testing of this reactor design to date took place during a seven-month period at the Texas Street Well Facility in Redlands, Calif. (*40*). Two rectangular upflow PBRs (2.1 m tall × 0.61 m wide × 0.30 m deep) were built at the site to treat groundwater containing ~80 µg/L of perchlorate. One reactor contained coarse sand as a packing material, and the other used a random plastic medium. Each reactor was inoculated with a pure culture of the perchlorate degrader *Azospira* sp. KJ and was fed acetate as a microbial growth substrate. At a flow rate of 3.8 L/min, the plastic-medium reactor consistently removed both perchlorate and nitrate to below detectable levels (4 µg/L and 0.1 mg/L, respectively). The sand reactor also removed both contaminants at this flow for short periods, but system performance then became inconsistent because of excessive biomass growth and channeling within the reactor. Instituting weekly backwashing cycles to remove biomass from the medium resolved the issue.

The PBR design has not yet been applied at full scale for perchlorate treatment, although tests with flow rates as high as 76 L/min have been conducted (*30*). All laboratory and pilot tests to date suggest that the PBR can effectively remove perchlorate to a nondetectable level. However, for this technology to be practical at full scale, a reliable method is needed to control the biomass inventory within the reactor over long periods while maintaining perchlorate levels below applicable standards. At flows of thousands of liters per minute, which are often required for groundwater treatment, it remains to be seen whether backwashing cycles or other methods will work.

FBRs are fixed-film reactors in which the biological medium is suspended or fluidized within the reactor vessel by the upward flow of water through the system (*41, 42*). Combining influent water with treated water recycled from the top of the reactor generates the flow. Although various materials have been tested, usually sand or granular activated carbon (GAC) is common in these systems as the microbial support medium. The small, suspended media particles provide a large surface area for microbial growth and promote a biomass density that is often several times that of other bioreactor designs, thus allowing a much higher volumetric efficiency (*42*). In addition, because of scouring from the fluidization process and the development of biomass control systems, these reactors do not typically clog like PBRs (*41–43*). The main disadvantage of this reactor design is that higher flow rates than in a PBR are necessary to maintain bed fluidization, and, subsequently, larger pumps and more energy are required for operation.

Four full-scale FBR systems—a total of 14 re-

actors—currently treat perchlorate in the United States, and one additional reactor is almost complete. The first full-scale system was built at the Aerojet facility in Rancho Cordova, Calif., in 1998, after extensive pilot-testing of different treatment approaches (*44*). This system consists of four FBRs, each measuring 4.3 m diam × 6.7 m high and containing ~18,000 kg of GAC media. The FBRs are fed ethanol as an electron donor and were seeded with waste solids from a food processing facility, which provided a microbial inoculum for perchlorate reduction. Since inoculation and start-up in 1998, these reactors have treated >30 billion L of groundwater, with influent levels ranging from ~2 to 7 mg/ L and effluent levels consistently below the MRL of 4 µg/L. The reactors now treat ~19,000 L/min (~27 million L/day) of groundwater. In 2001, the operation and maintenance costs for this reactor system were ~$0.20 per 3800 L of treated water (*45*).

Three additional full-scale FBR systems have been constructed since 1998: the Longhorn Army Ammunition Plant (Karnack, Texas; *46*), the Mc-Gregor Naval Weapons Industrial Reserve Plant (McGregor, Texas), and the Kerr–McGee production facility (Henderson, Nev.; shown in opening picture). Longhorn and McGregor have single reactors with influent flows of ~190 and 570 L/min, respectively. The influent concentrations of perchlorate to these reactors range from ~2.3 mg/L for the McGregor system to 25 mg/L for the Longhorn reactor. Each has consistently produced effluent water with perchlorate levels less than the MRL of 4 µg/L. The system in Henderson consists of 8 4.3-m-diam FBRs; 4 contain sand, and 4 use GAC as support media. These reactors are sequenced such that the influent water flows through the four sand-based FBRs followed by the four GAC-based reactors for polishing. This system is designed to treat ~3800 L/min of groundwater with perchlorate levels up to 400 mg/L in the influent. This system also treats chlorate and nitrate, which are commingled with perchlorate at the site.

Drinking-water treatment

With the increasing demand for potable water in the southwestern United States, particularly in southern California, biological removal of perchlorate and nitrate from drinking water is receiving serious consideration. For this approach to be viable, several issues must be considered, including water quality and safety, reliable system design and control, regulatory approval, and technology acceptance by drinking-water purveyors and the public at large. It is too early to predict the future of bioreactor-based drinking-water systems, but significant progress has been made during the past few years toward the development, testing, and regulatory approval of this technology.

Various designs of hydrogen-based reactors have been tested in laboratory studies for treating nitrate (e.g., *47*–*49*) and, more recently, chlorate (*50*) and perchlorate in drinking water (*51*–*53*). The expected advantage of these systems is the absence of any remaining organic electron donor in the treated water. Furthermore, biomass growth (and thus, total suspended solids in the effluent and the potential for reactor biofouling) tends to be less in such autotrophic systems than in heterotrophic ones that use organic electron donors.

The main drawback is that hydrogen is only sparingly soluble in water; this can result in an insufficient supply being available to completely bioreduce perchlorate, particularly in waters containing appreciable quantities of competing electron acceptors such as nitrate and oxygen (*51, 52*). In addition, hydrogen vented from these systems represents a potential explosion hazard. The use of hollow-fiber membranes (*53*) or electrolysis of water (*54*) to provide soluble hydrogen is likely to overcome the supply constraints and minimize the explosion potential associated with hydrogen-based bioreactors. However, the technical and economic viability of full-scale systems remains to be determined.

A conventional FBR that uses an organic substrate has also been evaluated as part of a treatment train to produce drinking water from perchlorate-impacted groundwater (*55*). During this study, one of the full-scale FBRs operating at the Aerojet facility removed perchlorate from groundwater as part of a drinking-water pilot demonstration. The drinking-water treatment system also included downstream oxygenation and filtration to remove residual ethanol and suspended solids, a liquid-phase GAC system, chlorine disinfection, and unit operations to treat organic co-contaminants (e.g., trichloroethene and *N*-nitrosodimethylamine) in the groundwater. The effluent from the treatment system was evaluated for a wide variety of water quality parameters for 1.5 yr at flow rates as high as 5300 L/min.
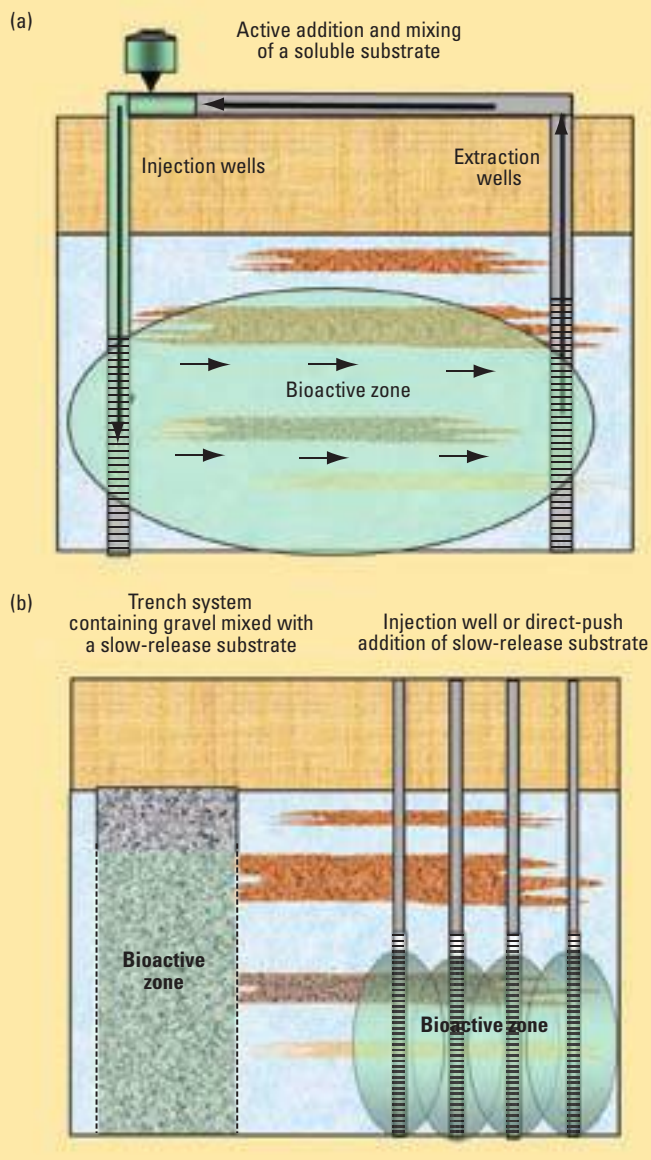
After reviewing the operating data, an expert panel concluded that the FBR is an effective technology for removing perchlorate from water and should be a reliable component of a treatment train for the production of potable water (*56*). On the basis of this study, DHS issued a letter in 2002 that provided conditional acceptance of FBRs for the removal of perchlorate during drinking-water production (*57*). This letter included several requirements, including the use of National Sanitation Foundation Standard 60 certified chemicals, on-line monitoring for nitrate and perchlorate, and downstream treatment equivalent to that used to produce drinking water from surface water.

Three years after the DHS approval, no biologically based drinking-water systems for perchlorate have been built in California. The slow implementation of this technology appears to reflect both the necessarily cautious nature of the drinking-water industry toward new treatment approaches and the relatively low perchlorate concentrations present in much of the water requiring treatment. When perchlorate concentrations are in the low-micrograms-per-liter range, ion-exchange approaches are much more cost-effective than at higher levels. In addition, ion exchange is widely accepted for drinking-water treatment; thus, this approach has been used for all of the major drinking-water applications to date. The long-term prospects for biologically based drinking-water systems will likely hinge upon some

FIGURE 2

In situ treatment of perchlorate can be done in several ways.

(a) A groundwater recirculation system designed to actively amend an aquifer with soluble substrate and (b) two approaches for adding slow-release substrates to the subsurface. Indigenous bacteria use the substrates to degrade perchlorate to chloride and oxygen.

(a)

Active addition and mixing of a soluble substrate

Injection wells

Extraction wells

Bioactive zone

(b)

Trench system containing gravel mixed with a slow-release substrate

Injection well or direct-push addition of slow-release substrate

Bioactive zone

Bioactive zone

mediation purposes. As previously noted, perchlorate-reducing bacteria occur widely in nature, so bioaugmentation in the field should be unnecessary (*22*, *23*, *26–28*). In addition, these organisms can often be stimulated to degrade perchlorate to below the current reporting limit of 4 μg/L through the addition of different electron donors, including ethanol, acetate, lactate, citrate, compost materials, polylactate ester, or vegetable oil (*16*, *17*, *26–28*, *30*). The most effective electron donors vary somewhat by site, and environmental conditions such as low pH and high salinity can impede biodegradation. However, most laboratory data suggest that in situ biostimulation through electron donor addition can be a viable remedial approach.

On the basis of promising laboratory studies, several field demonstrations of in situ perchlorate bioremediation have been conducted, and one full-scale biobarrier has been installed. The type of electron donor and the design of the electron-donor delivery system differentiate these in situ approaches. Two primary approaches have been tested for in situ perchlorate treatment. One is engineered systems that meter and mix soluble electron donors into groundwater, usually using a groundwater recirculation design; the other approach is to emplace slow-release electron donors in trenches or to use direct-push methods (Figure 2).

Slow-release materials are likely to be most useful and economically viable in shallow groundwater (treatment zone <15 m deep) and where perchlorate plumes are reasonably small. In deeper groundwater, the intensive drilling required to emplace and distribute these poorly dispersing materials is likely to be cost-prohibitive. Rather, recirculation systems, which can use a limited number of groundwater extraction and reinjection wells yet have a wide zone of influence, are likely to be more economical.

Three field demonstrations to evaluate the effectiveness of groundwater recirculation systems for perchlorate bioremediation have been completed, and several others are ongoing (*16*, *17*, *58*). In a study recently completed at the Naval Surface Warfare Center's Indian Head Division in Maryland, a recirculation system used a pair of extraction wells to intercept groundwater in a perchlorate source area. Then, an automated injection system amended that groundwater with the electron-donor lactate before reinjection to the subsurface (*58*; Figure 3). A carbonate/bicarbonate buffer was also added to the aquifer to increase pH, because microcosm studies suggest that low pH inhibits perchlorate degradation at this location even when appropriate electron donors are added. The average perchlorate concentration in the aquifer, which was initially >170 mg/L, declined by >95% in 8 of 9 monitoring wells during the 5-month demonstration period, with 5 wells reaching <1 mg/L and 2 dropping below the study's MRL of 5 μg/L.

An identical recirculation cell that received neither lactate nor buffer showed no appreciable reduction in perchlorate or nitrate levels during the test period. These data suggest that in situ perchlorate bioremediation using a recirculation design

combination of water purveyor acceptance, economic considerations, and the number and types of sources requiring treatment when regulations are firmly established.

In situ bioremediation

Various technologies have been developed to biologically treat groundwater contaminants in place. Previous approaches include biostimulation, whereby amendments are mixed with groundwater to promote contaminant degradation by indigenous microorganisms, and bioaugmentation, in which exogenous cultures are added to an aquifer for re-

can be a successful approach for treating perchlorate source areas.

A similar extraction–reinjection design was successfully tested as a cutoff barrier at the Aerojet site (*16*). After a small-scale pilot system showed promising results, a larger system was constructed that consisted of 2 extraction wells spaced ~60 m apart, with a single injection well centered between them. The system was designed to capture and treat a perchlorate plume that is ~180 m wide. In this case, groundwater was pumped from the two extraction wells to the surface, where it was amended with an electron donor (ethanol or citric acid) and then recharged to the aquifer via the single recharge well. During the demonstration, perchlorate concentrations were consistently reduced from ~8 mg/L to <4 µg/L within ~10 m of the injection well.

Other field demonstrations using soluble electron donors with various injection and mixing strategies, including recirculation cells, semipassive barriers, and horizontal flow treatment wells, are ongoing in California, Utah, Nevada, Texas, and Maryland. A demonstration under way at the Longhorn site in Texas uses a series of sequenced injection and extraction wells to mix lactate into a perchlorate plume emanating from a closed landfill. Unlike previous field trials, the Texas wells are pumped only intermittently to achieve mixing of the electron donor within a specified cutoff zone. This semipassive approach maximizes substrate distribution while minimizing the operational costs and maintenance issues associated with active pumping systems (*59*).

FIGURE 3

A field pilot system used to test the effectiveness of in situ perchlorate treatment at a U.S. Navy site.

(a) In a study recently completed in Indian Head, Md., a recirculation system with a pair of extraction wells was used to intercept groundwater in a perchlorate source area. Then, prior to reinjection to the subsurface, an automated injection system amended that groundwater with the electron donor lactate. Perchlorate data from (b) shallow and (c) deep monitoring wells show good performance.
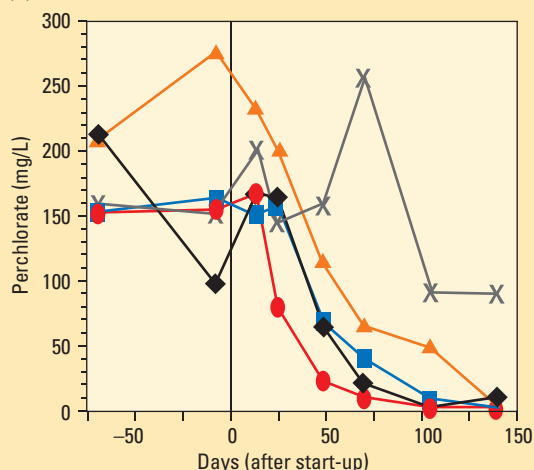
(a)
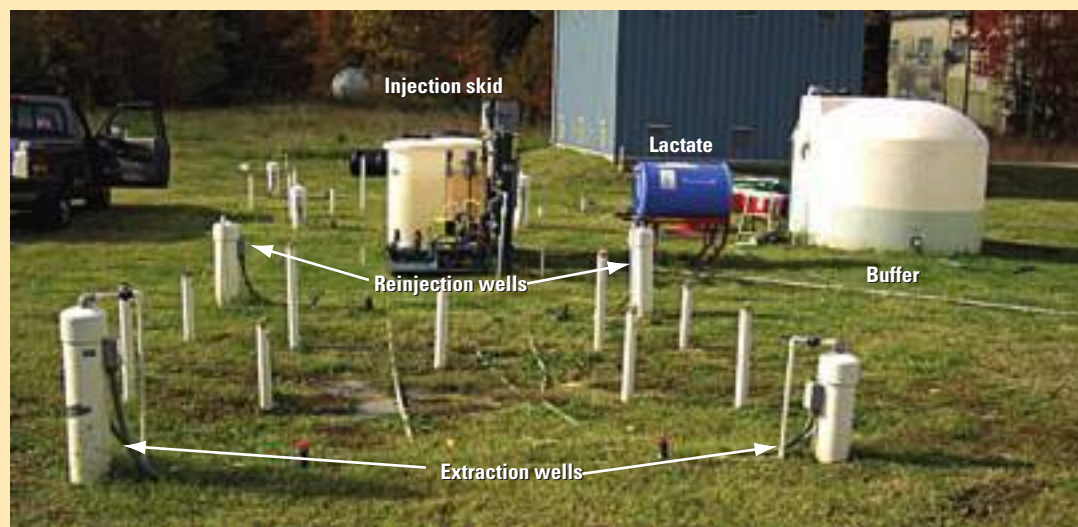
(b)
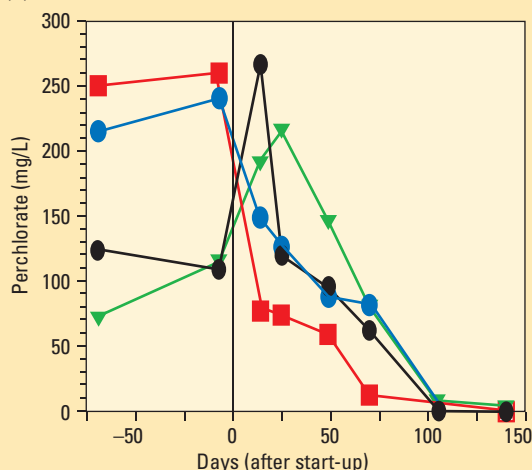
(c)

Slow-release electron donors, such as vegetable oils, compost materials, pecan shells, and wood chips, have been shown in laboratory and in recent pilot demonstrations to support perchlorate bio-degradation (*26, 60–62*). In a Maryland field study, the passive injection of emulsified vegetable oil was reported to promote the biodegradation of both perchlorate and 1,1,1-trichloroethane in a shallow aquifer (*62*). Perchlorate levels declined from ~9000 mg/L to <10 µg/L in down-gradient monitoring wells within 35 days of substrate injection. In another field demonstration, pecan shells and cottonseed were used as part of an in situ barrier to stimulate bioreduction of perchlorate and nitrate discharged by a radioactive waste treatment facility at Los Alamos National Laboratory (*63*). This barrier, which is ~7 m across and 8 m deep, also contains layers of permeable materials, including gravel to trap colloids and apatite mineral to remove dissolved radionuclides and metals. Initial monitoring results reveal that perchlorate is degraded in both the apatite and pecan-shell layers. However, drought conditions have limited the amount of groundwater moving through the system during the past year.

To date, the only full-scale in situ remediation system designed specifically for perchlorate treatment is located at the site in McGregor, Texas, and consists of a series of 7 shallow trenches (~3–6 m deep) totaling >1000 m in length (*64*). These trenches are filled with gravel and a mixture of mushroom compost and soybean-oil-soaked wood chips, which are used as slow-release electron substrates. The trenches, which serve as a series of cut-off barriers for groundwater and surface water at the site, also include internal piping that can be used to apply a soluble electron donor to further enhance the rates of perchlorate reduction as necessary on the basis of effluent perchlorate levels. As contaminated water flows through the trenches, perchlorate is biologically degraded, preventing off-site migration. Data from the project show that influent perchlorate concentrations as high as 13 mg/L are being reduced to below detection levels.

Future of bioremediation

It may take several years to develop and implement a federal drinking-water standard for perchlorate. Until then, or until state recommendations become law rather than guidance or action levels, the scope and timing of perchlorate remediation remain unclear. If regulatory levels are set in the low-micrograms-per-liter range, as most current state advisory levels are, the total extent of cleanup could be enormous. If, however, a much higher drinking-water standard is set, future efforts may focus primarily on cleaning up point sources at military, aerospace, and industrial facilities. In either case, on the basis of the economics and reliability of full-scale bioreactor systems and the encouraging field data from numerous in situ approaches, biological treatment is likely to play a prominent role as a remedial solution for perchlorate in the future.

SAM FRISCH

The Longhorn Army Ammunition Plant (Karnack, Texas).

Paul B. Hatzinger is a senior research scientist with Shaw Environmental, Inc., in Lawrenceville, N.J. Address correspondence regarding this article to paul.hatzinger@shawgrp.com.

References

(1) California Department of Health Services. *Perchlorate in California Drinking Water: Monitoring Update*, www.dhs.ca.gov/ps/ddwem/chemicals/perchl/monitoringupdate.htm.

(2) U.S. EPA, Federal Facilities Restoration and Reuse. *National Perchlorate Detections as of September 23, 2004*, www.epa.gov/fedfac/documents/perchlorate_map/nationalmap.htm.

(3) Hogue, C. Rocket-Fueled River. *Chem. Eng. News* **2003**, *81*, 37–46.

(4) Massachusetts Military Reservation Impact Area Groundwater Study Program, www.groundwaterprogram.org.

(5) Urbansky, E. T.; et al. Perchlorate Levels in Samples of Sodium Nitrate Fertilizer Derived from Chilean Caliche. *Environ. Pollut.* **2001**, *112*, 299–302.

(6) Orris, G. J.; et al. *Preliminary Analyses for Perchlorate in Selected Natural Materials and Their Derivative Products*; Open-File Report 03-314; U.S. Geological Survey: Tucson, AZ, 2003.

(7) Christen, K. Perchlorate Mystery Surfaces in Texas. *Environ. Sci. Technol.* **2003**, *37*, 376A–377A.

(8) U.S. Food and Drug Administration, Exploratory Data on Perchlorate in Food, http://vm.cfsan.fda.gov/~dms/clo4data.html.

(9) *Perchlorate Environmental Contamination: Toxicological Review and Risk Characterization*. External Review Draft, NCEA-1-0503; U.S. EPA Office of Research and Development, National Center for Environmental Assessment: Washington, DC, 2002.

(10) National Research Council. *Health Implications of Perchlorate Ingestion*. The National Academies Press: Washington, DC, 2005.

(11) Pleus, R. C. Perchlorate: Using Good Science to Derive a Safe Drinking Water Level. *Water Cond. Purif.* **2003**, *45* (8), 36–40.

(12) Wang, H.-C.; Eaton, A.; Narloch, B. *National Assessment of Perchlorate Contamination Occurrence*; American Water Works Association: Denver, CO, 2002.

(13) California Department of Health Services, www.dhs.ca.gov/ps/ddwem/chemicals/perchl/perchlorateforweb.xls.

(14) Logan, B. E. Assessing the Outlook for Perchlorate Remediation. *Environ. Sci. Technol.* **2001**, *35*, 483A–487A.

(15) Damian, P.; Pontius, F. W. From Rockets to Remediation: The Perchlorate Problem. *Environ. Protect.* **1999**, *June*, 24–31.

(16) Cox, E. E.; McMaster, M.; Neville, S. L. Perchlorate in Groundwater: Scope of the Problem and Emerging Re-

medial Solutions. In *Proceedings of the 36th Annual Engineering Geology and Geotechnical Engineering Symposium*; Las Vegas, NV, 2001; pp 27–32.

(17) Hatzinger, P. B.; et al. In-Situ and Ex-Situ Bioremediation Options for Treating Perchlorate in Groundwater. *Remediation* **2002**, *12*, 69–85.

(18) Boodoo, F. POU/POE Removal of Perchlorate. *Water Cond. Purif.* **2003**, *45* (8), 52–55.

(19) Urbansky, E. T., Ed. *Perchlorate in the Environment*; Kluwer Academic/Plenum Publishers: New York, 2000.

(20) Achenbach, L. A.; et al. *Dechloromonas agitata* gen. nov., sp. nov. and *Dechlorosoma suillum* gen. nov., sp. nov., Two Novel Environmentally Dominant (Per)chlorate-Reducing Bacteria and Their Phylogenetic Position. *Int. J. Syst. Evol. Microbiol.* **2001**, *51*, 527–533.

(21) Zhang, H.; Bruns, M. A.; Logan, B. E. Perchlorate Reduction by a Novel Chemolithoautotrophic, Hydrogen-Oxidizing Bacterium. *Environ. Microbiol.* **2002**, *4*, 570–576.

(22) Coates, J. D.; et al. Ubiquity and Diversity of Dissimilatory (Per)chlorate-Reducing Bacteria. *Appl. Environ. Microbiol.* **1999**, *65*, 5234–5241.

(23) Coates, J. D.; Achenbach, L. A. Microbial Perchlorate Reduction: Rocket-Fuelled Metabolism. *Nature Rev. Microbiol.* **2004**, *2*, 569–580.

(24) Song, Y.; Logan, B. E. Effect of O_2 Exposure on Perchlorate Reduction by *Dechlorosoma* sp. KJ. *Water Res.* **2004**, *38*, 1626–1632.

(25) Chaudhuri, S. K.; et al. Environmental Factors That Control Microbial Perchlorate Reduction. *Appl. Environ. Microbiol.* **2002**, *68*, 4425–4430.

(26) Wu, J.; et al. Persistence of Perchlorate and the Relative Numbers of Perchlorate- and Chlorate-Respiring Microorganisms in Natural Waters, Soils, and Wastewater. *Biorem. J.* **2001**, *5*, 119–130.

(27) Waller, A. S.; Cox, E. E.; Edwards, E. A. Perchlorate-Reducing Microorganisms Isolated from Contaminated Sites. *Environ. Microbiol.* **2004**, *6*, 517–527.

(28) Tipton, D. K.; Rolston, D. E.; Scow, K. M. Transport and Biodegradation of Perchlorate in Soils. *J. Environ. Qual.* **2003**, *32*, 40–46.

(29) Rikken, G. B.; Kroon, A. G. M.; van Ginkel, C. G. Transformation of (Per)chlorate into Chloride by a Newly Isolated Bacterium: Reduction and Dismutation. *Appl. Microbiol. Biotechnol.* **1996**, *45*, 420–426.

(30) Xu, J. ; et al. Microbial Degradation of Perchlorate: Principles and Applications. *Environ. Eng. Sci.* **2003**, *20*, 405–422.

(31) Kengen, S. W. M.; et al. Purification and Characterization of (Per)chlorate Reductase from the Chlorate-Respiring Strain GR-1. *J. Bacteriol.* **1999**, *181*, 6706–6711.

(32) van Ginkel, C. G.; et al. Purification and Characterization of Chlorite Dismutase: A Novel Oxygen-Generating Enzyme. *Arch. Microbiol.* **1996**, *166*, 321–326.

(33) Rittman, B. E.; McCarty, P. L. *Environmental Biotechnology: Principles and Applications*; McGraw-Hill: New York, 2001.

(34) Attaway, H.; Smith, M. D. Propellant Wastewater Treatment Process. U.S. Patent 5,302,285, 1994.

(35) Wallace, W.; et al. Identification of an Anaerobic Bacterium Which Reduces Perchlorate and Chlorate as *Wolinella succinogenes*. *J. Ind. Microbiol.* **1996**, *16*, 68–72.

(36) *Ammonium Perchlorate Biodegradation for Industrial Wastewater Treatment*; Cost and Performance Report CP-9710; Environmental Security Technology Certification Program: Arlington, VA, 2000.

(37) Wallace, W.; et al. Perchlorate Reduction by a Mixed Culture in an Up-Flow Anaerobic Fixed Bed Reactor. *J. Ind. Microbiol. Biotechnol.* **1998**, *20*, 126–131.

(38) Kim, K; Logan, B. E. Fixed-Bed Bioreactor Treating Perchlorate-Contaminated Waters. *Environ. Eng. Sci.* **2000**, *17*, 257–265.

(39) Losi, M. E.; et al. Bioremediation of Perchlorate-Contaminated Groundwater Using a Packed Bed Biological Reactor. *Biorem. J.* 2002, *6*, 97–103.

(40) Min, B.; et al. Perchlorate Removal in Sand and Plastic Media Bioreactors. *Water Res.* **2004**, *38*, 47–60.

(41) Sutton, P. M.; Mishra, P. N. Activated Carbon Based Biological Fluidized Beds for Contaminated Water and Wastewater Treatment: A State-of-the-Art Review. *Water Sci. Technol.* **1994**, *29*, 309–317.

(42) *Nitrogen Control Manual*; EPA/625/R-93/010; U.S. EPA Office of Research and Development: Washington, DC, 1993.

(43) Frisch, S. Bed Cleaning System for Fluidized Bed Bioreactors. U.S. Patent 6,706,421B2, 2004.

(44) Hatzinger, P. B.; et al. Treatment of Perchlorate-Contaminated Groundwater Using Fluidized Bed Reactors. In *Case Studies in the Remediation of Chlorinated and Recalcitrant Compounds*; Wickramanayake, G. B., et al., Eds.; Battelle Press: Columbus, OH, 2001; pp 115–122.

(45) U.S. EPA, Aerojet Corp. documents, http://yosemite.epa.gov/r9/sfund/TECHdoc.nsf/0/44b7ec34f30dc8b888256bd7005767ee?OpenDocument.

(46) Polk, J.; et al. Army Success Story: Ex-Situ Biological Treatment of Perchlorate-Contaminated Groundwater. *Fed. Facil. Environ. J.* **2002**, *13*, 85–94.

(47) Kurt, M.; Dunn, I. J.; Bourne, J. R. Biological Denitrification of Drinking Water Using Autotrophic Organisms with H_2 in a Fluidized-Bed Biofilm Reactor. *Biotechnol. Bioeng.* **1987**, *29*, 493–501.

(48) Delanghe, B.; et al. Drinking Water Denitrification in a Membrane Bioreactor. *Water Sci. Technol.* **1994**, *30*, 157–160.

(49) Lee, K.-C.; Rittmann, B. E. Applying a Novel Autohydrogentrophic Hollow-Fiber Membrane Biofilm Reactor for Denitrification of Drinking Water. *Water Res.* **2002**, *36*, 2040–2052.

(50) Kroon, A. G. M.; van Ginkel, C. G. Biological Reduction of Chlorate in a Gas-Lift Reactor Using Hydrogen as an Energy Source. *J. Environ. Qual.* **2004**, *33*, 2026–2029.

(51) Logan, B. E.; LaPoint, D. Treatment of Perchlorate- and Nitrate-Contaminated Groundwater in an Autotrophic, Gas Phase, Packed-Bed Bioreactor. *Water Res.* **2002**, *36*, 3647–3653.

(52) Miller, J. P.; Logan, B. E. Sustained Perchlorate Degradation in an Autotrophic, Gas-Phase, Packed-Bed Bioreactor. *Environ. Sci. Technol.* **2000**, *34*, 3018–3022.

(53) Nerenberg, R.; Rittmann, B. E.; Najm, I. Perchlorate Reduction in a Hydrogen-Based Membrane-Biofilm Reactor. *J. Amer. Water Works Assoc.* **2002**, *94*, 103–114.

(54) Sakakibara, Y.; et al. Denitrification and Neutralization with an Electrochemical and Biological Reactor. *Water Sci. Technol.* **1994**, *30*, 151–155.

(55) Phase 2 Treatability Study Report—Aerojet GET E/F Treatment Facility, http://clu-in.org/download/contaminant focus/perchlorate/Phase_2_TS-September_2001.pdf.

(56) *Review of Phase 2 Treatability Study, Aerojet Facility, Rancho Cordova,* California, www.clu-in.org/download/contaminantfocus/perchlorate/Phase_2_TS-September_2001.pdf.

(57) California Department of Health Services letter, www.safedrinkingwater.com/community/Aerojetletter040222.pdf.

(58) Cramer, R. J.; et al. *Field Demonstration of In Situ Perchlorate Bioremediation at Building 1419.* Naval Ordnance Safety and Security Activity, Naval Sea Systems Command Technical Report No. NOSSA-TR-2004-001, Indian Head, MD, 2004.

(59) Environmental Security Technology Certification Program, Remediation Projects, www.estcp.org.

(60) Perlmutter, M. W.; et al. Innovative Technology: In Situ Bioremediation of Perchlorate-Contaminated Groundwater. In *Proceedings Air and Waste Management Association, 93rd Annual Conference and Exhibition*; Salt Lake City, UT, 2000.

(61) Hunter, W. J. Bioremediation of Chlorate or Perchlorate Contaminated Water Using Permeable Barriers Containing Vegetable Oil. *Cur. Microbiol.* **2002**, *45*, 287–292.

(62) Zawtocki, C.; Lieberman, T. M.; Birk, G. A Dash of Oil and Let Marinade. *Pollut. Eng.* **2004**, *May*, 30–34.

(63) Geochemical and Biogeochemical Mechanisms of Contaminant Attenuation in a Multi-Layered Permeable Reactive Barrier, http://gsa.confex.com/gsa/2003AM/finalprogram/abstract_67692.htm.

(64) U.S. EPA, Biological PRB Used for Perchlorate Degradation in Ground Water, www.clu-in.org/products/newsltrs/tnandt/view.cfm?issue=0204.cfm#2.