

United States
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Bioremediation: Innovative Pollution Treatment Technology

A Focus on EPA's Research



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EPA's Office of Research and Development

The Office of Research and Development (ORD) conducts an integrated program of scientific research and development on the sources, transport and fate processes, monitoring, control, and assessment of risks and effects of environmental pollutants. These activities are implemented through its headquarters offices, technical support offices, and twelve research laboratories distributed across the country. Research focuses on key scientific and technical issues to generate knowledge supporting sound decisions today and anticipating the complex challenges of tomorrow. With a strong, forward-looking research program, less expensive, more effective solutions can be pursued and irreversible damage to the environment prevented.

*Front Cover:
Bioremediation **has been an**
effective **treatment technique** for the
reclamation of **ocean beaches**
contaminated as **a result of crude oil**
spills –*

Alan Pitcairn/Grant Heilman Photography Photo

The United States is the world leader in the field implementation of bioremediation, an attractive alternative to conventional methods of cleaning up persistent hazardous wastes in the environment.

Complex synthetic chemicals and petroleum derivatives have accumulated in the environment as waste materials for decades. Conventional treatments, such as excavation followed by incineration, have been used for some time to clean hazardous waste sites, but can be costly and inherently disruptive to the environment. The United States Environmental Protection Agency's (EPA's) Office of Research and Development plays a major role in the basic science and engineering involved in developing and supporting innovative technologies that are cost-effective alternatives to existing methods for cleaning up hazardous waste sites and oil spills. One of the most promising of these new methods for solving toxic waste cleanup problems is bioremediation.

Bioremediation technology can be a non-disruptive, cost-effective, and highly efficient method of destroying many environmentally persistent toxic chemicals. Although the development of bioremediation has progressed rapidly over the past several years, a great deal must still be accomplished before the technology can be fully utilized. This is true in terms of scientific research, the engineering design of treatment systems, and field evaluations. In response to these needs, ORD has developed an integrated Bioremediation Program to advance the understanding,

development, and application of bioremediation technologies to help solve hazardous waste problems threatening human health and the environment. As these technologies advance, ORD transfers information on their use to groups who apply them to treat specific sites.

What Bioremediation Involves

Bioremediation technologies typically use naturally occurring microorganisms (bacteria or fungi) to degrade hazardous wastes. Like all living creatures, microbes need nutrients, carbon, and energy to survive and multiply. Such organisms are capable of breaking down toxic chemicals to obtain food and energy, typically degrading them into harmless substances consisting

Electron micrograph of Pseudomonas aeruginosa, oil-degrading bacteria (magnification at X5,500).



Manfred Kage/Peter Arnold, Inc. Photo



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After the target chemical has been biodegraded, most of the microbial population will die off naturally since there will no longer be a sufficient food or energy source for the microorganisms to survive.

Two Alaskan beach field test plots show that a site where fertilizer was applied is much cleaner than a site where no fertilizer was added.

mainly of carbon dioxide, water, salts, other innocuous products, and sometimes methane.

Microorganisms are present everywhere in nature, even in the deep ocean, and are an integral part of the earth's natural detoxification process. Bioremediation technologies harness this process by promoting the growth of competent populations of microorganisms that can biodegrade contaminants.

Biostimulation and Bioaugmentation

Some microbes capable of degrading target contaminants are often already present at a hazardous waste site, although not necessarily in the numbers required to remediate the site. In these cases, methods are devised to stimulate the growth and biodegradative activities of the existing microbial communities. Such biostimulation usually involves adding nutrients or oxygen to the contaminated material to help the indigenous microorganisms flourish. The greater the population of degrading microorganisms within the controlled remediation area, the faster

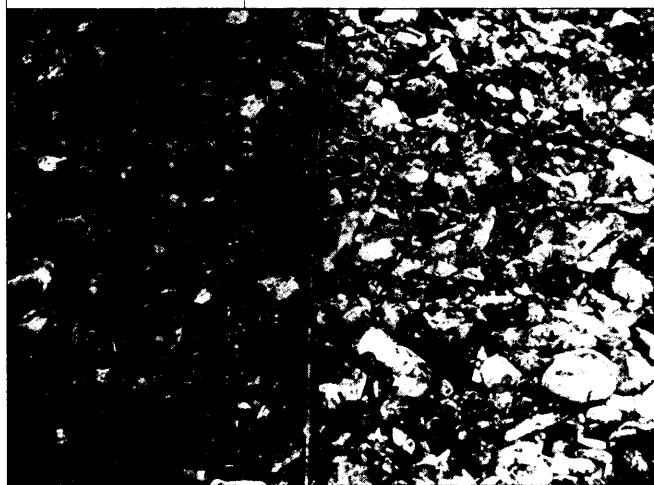
and more efficient the biodegradation process. At present, most sites being treated with bioremediation use indigenous microorganisms.

During the summer following the 1989 Exxon Valdez oil spill in Alaska's Prince William Sound, ORD initiated a bioremediation field demonstration to determine the feasibility of using nutrients to stimulate the indigenous microbial degradation of oil on the Alaskan shoreline.

The project involved applying fertilizers containing nitrogen and phosphorus (nutrients bacteria need to utilize crude oil hydrocarbons as a food source) to selected test plots on oil-covered cobblestone and sand and gravel beaches. Within two weeks after applying the fertilizer to the test plots, scientists began to observe reductions in the amount of oil on treated beach surfaces. Non-treated plots remained as oiled as they had been at the beginning of the field study.

During the demonstration, several sampling and field testing methods were used to observe changes in the composition of the oil, monitor the movement of added nutrients on the test beaches, detect changes in the number of bacteria present, and assess the degradation of the oil. This ORD study, which was the largest project of its kind ever conducted, clearly demonstrated the capabilities of biostimulation techniques to remediate oil spills in the field. It has also provided a wealth of data that will have far-reaching implications for successfully mitigating the effects of future oil spills worldwide.

In cases where insufficient indigenous microorganisms are present at a site to degrade the target hazardous wastes even with



biostimulation, non-indigenous microorganisms known to metabolize the pollutants can be added to the affected material. Adding species that are known to work in concert with resident microorganisms (bioaugmentation) can result in faster or more complete waste degradation.

For example, one ORD study conducted by EPA's Environmental Research Laboratory in Gulf Breeze, Florida, demonstrated the ability of selected non-indigenous microorganisms to facilitate biodegradation of ground water contaminated with creosote and pentachlorophenol (PCP). The ground water was taken from the American Creosote Works Superfund site in Pensacola, Florida. Results obtained from the addition of non-indigenous microorganisms were compared to those obtained using only indigenous organisms. During the study, more than 99% of creosote constituents in the samples and 87% of PCP were removed by the non-indigenous bacteria. When indigenous organisms were used alone, biodegradation was much less successful.

Bioremediation Potential

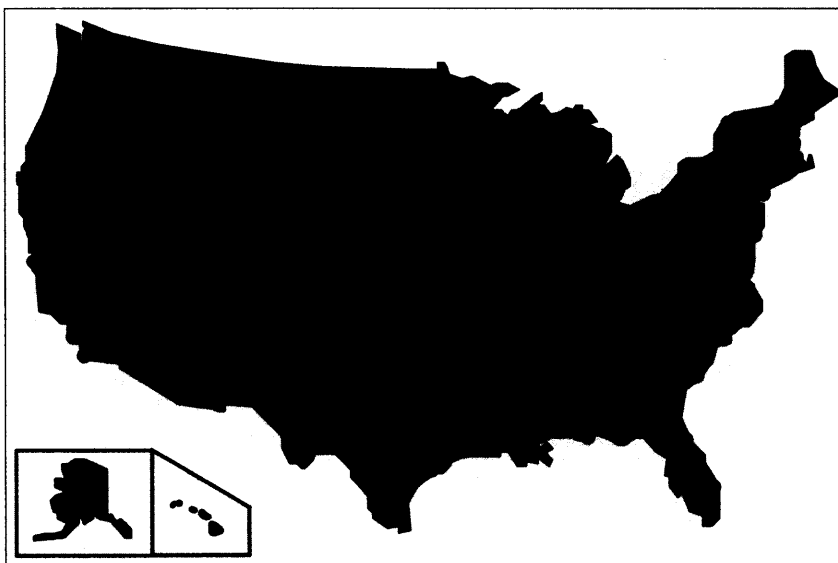
The potential use of bioremediation technologies is significant, as federal and state governments, private industry, and others responsible for environmental cleanup efforts add it to their arsenals

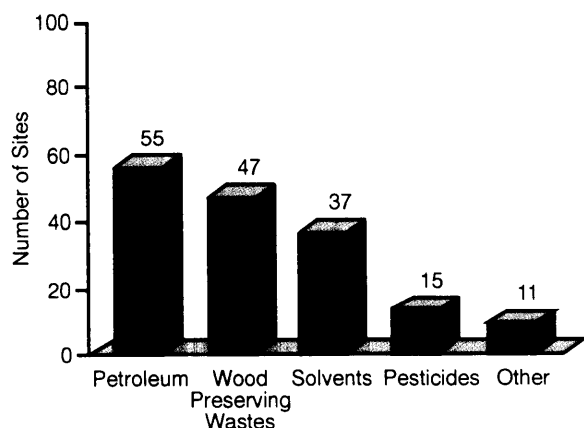
of methods for environmental reclamation.

To date, bioremediation projects are in planning stages, undergoing treatability studies, or in full-scale operation under federal or state regulatory authority at more than 150 sites across the United States. These include sites identified for cleanup under the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA, otherwise known as Superfund), the Resource Conservation and Recovery Act (RCRA), the Toxic Substances Control Act (TSCA), and Underground Storage Tank (UST) regulations. Of the approximately 1,240 National Priority List (NPL) sites already identified for cleanup, many are possible candidates for bioremediation.

An estimated fifteen percent of the nation's four to five million underground storage tanks containing petroleum, heating oil, and other hazardous materials are leaking, contaminating the soil around them and threatening or already contaminating ground water supplies. As many as

Bioremediation projects are being studied, planned, or are already implemented in thirty-six states.





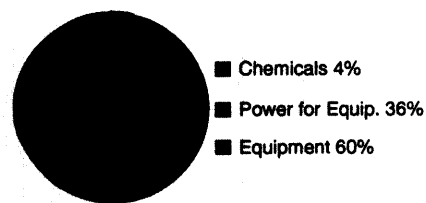
Various types of contamination are being treated using bioremediation at more than 150 sites under federal or state regulatory authority.

15,000 oil spills occur each year requiring cleanup of contaminated soils and waters. Additionally, thousands of RCRA facilities are contaminated with solvents, wood preservatives, halogenated aromatic hydrocarbons (HAHs), and pesticide wastes. And more than 10,000 pesticide dealerships throughout the country evidence contamination of soil and/or ground water. Bioremediation can play a significant role in the remediation of many of these sites.

Advantages

Bioremediation has some significant advantages when compared to other remediation technologies. The most important is the ability of microorganisms to detoxify hazardous substances instead of merely trans-

Typical operational expenses (without operator labor).



Source: Nyer, E. K. Groundwater Treatment Technology, Van Nostrand Reinhold Company, Inc. New York.

ferring contaminants from one environmental medium to another (such as from water to the air during air stripping).

Another major advantage, particularly for *in situ* (in place) treatment of soils, sludges, and ground water, is that bioremediation is usually less disruptive to the environment than other technologies used to remediate hazardous wastes, such as excavation followed by incineration and landfilling. And since treatment is normally accomplished on site, there is typically no need to transport hazardous materials to another location.

Finally, the cost of treating a hazardous waste site using bioremediation technologies can be considerably lower than with other treatment methods. For example, the cost of soil bioventing (discussed later) by a field-scale system has been estimated at less than \$50 per ton, while incineration costs are typically more than ten times that amount.

Limitations

The use of bioremediation is limited by the need for a greater understanding of biodegradation processes, their appropriate applications, their control and enhancement in the environment, and engineering techniques required for broader application of the technology. The EPA recognizes that comprehensive mechanistic process control, engineering design, and cost data are also necessary for the full acceptance and use of bioremediation by the technical and regulatory communities.

Bioremediation Involves More than Microbes

Although using microorganisms to degrade hazardous wastes may

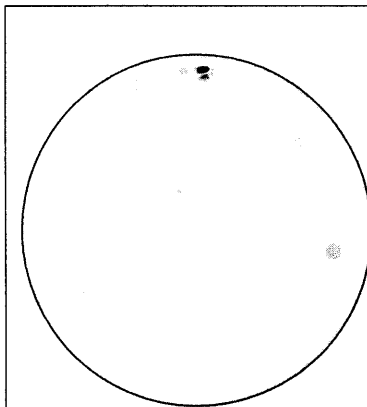
seem fairly straightforward, the technology of bioremediation is actually multifaceted and complex. It is based on extensive research to determine the biochemical capabilities of specific microorganisms to interact with specific waste compounds to successfully degrade them.

A great deal of information is also required about the characteristics of the individual waste site, including types and distribution of contaminants, properties of the contaminated media (soil, sediments, water, sludge), numbers and species of indigenous microorganisms, and topographical and subsurface geological properties. Once all of this is known, and it has been determined that the site is biologically treatable, a remediation plan is devised and engineering systems designed and implemented to accommodate the remediation process and measure the results.

The study of bioremediation requires a hybrid of several scientific and technical disciplines, including microbiology, ecology, biochemistry, analytical chemistry, chemical engineering, environmental engineering, geology, mathematics, statistics, civil engineering, and risk management. The EPA applies expertise in all of these fields to enhance the capabilities of the technology and match its promise as a major factor in decontaminating hazardous waste sites worldwide.

Ability of Microorganisms to Degrade Wastes

Individual strains of microbes have the capacity to degrade only



An important part of bioremediation is identifying the various microbial species present at a site. A preliminary step in this process can be to determine the total number of strains present by placing diluted sample material from the site onto an enriched culture medium. Different colors appearing on the culture (white, yellow, and orange in photo) indicate the presence of colonies of different strains. Colonies can then be collected from the culture and subjected to biochemical screening tests to identify the individual strains.

certain types of compounds. So, while a given species may very effectively degrade one compound, it may have no ability to degrade another. For this reason, understanding the specific metabolic capabilities of individual species is important to effectively match the right microorganisms or group of microorganisms with the target compound to be remediated.

Bacteria are, on average, one to two micrometers (millionths of a meter) in length. At this size, they interact with hazardous waste compounds on the molecular scale. Metabolism occurs when the microorganisms make contact with compound molecules and separate and absorb those useful for their nutritional and energy needs. Thus, microorganisms can degrade a toxic compound by systematically dismantling and consuming individual components of its molecular structure until there is nothing left but carbon dioxide, water, and other innocuous products.

The breakdown and digestion of waste compounds during the metabolic process are the result of biochemical reactions catalyzed by enzymes produced by the microor-

ganisms. Enzymes, which are complex proteins, are highly specific in their catalytic behavior, so a given enzyme is effective for only a particular type of chemical reaction. Because of this, the ability of a microorganism to degrade a particular substance depends upon its ability to produce enzymes capable of catalyzing the necessary biochemical reactions. ORD conducts ongoing research to identify additional enzyme systems and to characterize the full range of activities of enzymes already identified for use in bioremediation applications.

EPA research on microbial metabolic processes also provides important information about the types of additional nutrients and energy sources that individual strains require for growth, cell division (reproduction), and metabolism to biodegrade hazardous wastes.

Factors Limiting Biodegradation

While microorganisms are often described as microscopic biochemical reactors, their activities are intimately connected to and shaped by their external environment. Because

of this, any number of environmental conditions can slow or stop a biodegradation process even when the microorganisms have the ability to otherwise degrade the target compound. For example, the contaminated area may be too acidic or alkaline or the moisture conditions unfavorable for sufficient microbial metabolic activity to occur. Some microorganisms require the presence of oxygen to live (aerobic), while others live only in the absence of free oxygen (anaerobic). In other cases, the concentration of the target waste compound may be so high in the treatment area that it is toxic to the microorganisms.

ORD research on the external physical and chemical factors influencing microbial metabolism and growth is critical for developing efficient and cost-effective bioremediation technologies. A thorough understanding of such factors is important for creating optimum environmental conditions to stimulate the metabolic activities of microbial communities to degrade toxic wastes.

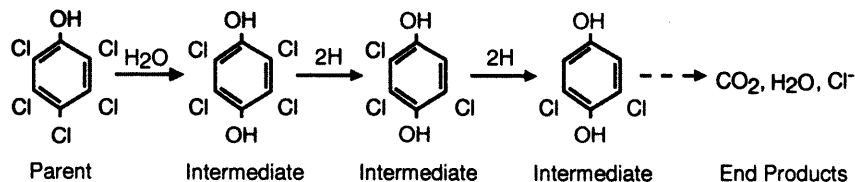
Before they can design proper treatment techniques, scientists and engineers must first determine which

Bioremediation rate-limiting factors are different for a porous beach environment than for a wetland or marsh.



Grant Heilman/Grant Heilman Photography Photo

The parent compound of a PCP congener is comprised of five chlorine molecules (Cl) and one hydroxide molecule (OH) in a carbon ring structure. In biodegradation, certain aerobic microbes break down the ring by metabolizing selected individual chlorine molecules, resulting in intermediate products and ultimately carbon dioxide, water, and chloride.



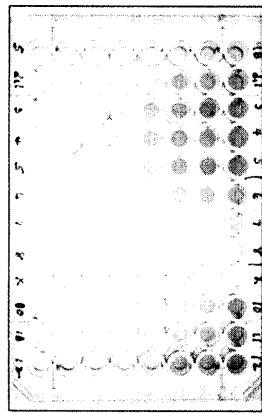
factors at a site would likely limit the rate and extent of biodegradation. The biodegradation of oil, for example, requires significant concentrations of nutrients and oxygen to proceed at a rate useful for bioremediation. The effects of these factors on oil biodegradation are quite different in a porous beach environment than a wetland or marsh. On a porous beach, which is constantly exposed to high concentrations of dissolved oxygen from tidal flows, oil biodegradation is likely to be limited by an insufficient supply of nutrients. A marsh, which is usually rich in organic carbon and nutrients, is likely to be limited by insufficient oxygen. Designing laboratory studies and microcosm systems to study methods for optimizing the rate and extent of biodegradation are important and cost-effective components of this ORD research.

Biodegradation Pathway

Microorganisms do not consume all of the digestible molecular constituents of a toxic compound at once. Instead, they selectively remove and metabolize individual components until a nondegradable product is formed or the compound has been completely degraded. Each time a molecular component is removed, the nature of the compound

The complexity of the biodegradation process necessitates innovative tools to accurately and cost-effectively assess any health and environmental impacts during and after bioremediation treatment. Scientists from ORD's Health Effects Research Laboratory in Research Triangle Park, North Carolina, and the Environmental Monitoring Systems Laboratory in Cincinnati, Ohio, are developing rapid, novel bioassays to estimate toxicity to humans and animals without the need for extensive chemical analysis. The Environmental Research Laboratory at Gulf Breeze, Florida, is performing similar studies to assess potential toxic effects on ecological systems.

Bioassays can be used to examine the effects of any intermediate and end products produced as a result of biodegradation when specified microorganisms are grown on the waste material of interest in a culture medium. Scientists can use these tests to accurately detect such toxicological properties as carcinogenicity and mutagenicity. The estimation of toxicity through the use of bioassays provides a powerful, cost-effective tool for the development of bioremediation applications that protect human health and the environment.



Some bioassays test the possible mutagenicity of target chemicals using selected indicator bacteria. If a chemical is mutagenic to these organisms, there is a probability that it could also be mutagenic or carcinogenic to humans and animals. In one bioassay developed by ORD, a chemical testing positive (mutagenic) is indicated by the formation of o-nitrophenol which produces a yellow color. The more intense yellow indicates a higher degree of DNA damage. No color is produced from chemicals testing negative.

is altered, resulting in the formation of a new substance. Consequently, metabolism involves a succession of new substances being formed (intermediates), beginning with the parent (original) and ending with the final product (end product). This is known as the biodegradation pathway.

An important concern of EPA's bioremediation research is understanding the chemical and biochemical reactions or pathways occurring during microbial degradation of hazardous waste compounds. This allows scientists to ensure that the intermediate and end products of the metabolic process are not more toxic than the original pollutant. Research has shown that intermediates and end products of biodegradation are most often less toxic than the parent compound.

Biodegradation by Microbial Communities

Some toxic compounds resistant to complete biodegradation by one strain of microorganism may be completely metabolized by a number of species working in concert. For example, one species may have the enzymatic machinery to only par-

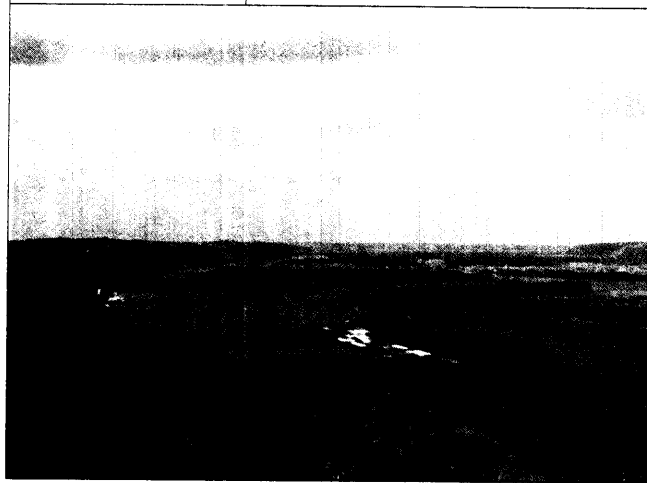
tially metabolize the parent compound, resulting in an intermediate product. Another species may be able to metabolize the intermediate product of the first species but lack the enzymes needed to metabolize the parent compound.

By themselves, neither species could totally degrade the toxic substance. But the combined metabolic activity of the two results in successful degradation of the compound. Microbial consortia consisting of two or more strains of microorganisms are typically required for the degradation of hazardous wastes.

ORD research in this area is directed toward identifying participating microbial species, the interactive and sequential roles played by the microorganisms, and any solubilizing agents they may produce to facilitate biodegradation. A more thorough understanding of the biochemistry, physiology, and ecology of these systems will lead to additional capabilities using consortia and sequential treatments to detoxify hazardous compounds.

The coupling of anaerobic dechlorination with aerobic metabolism has been suggested as a possible method for reducing the levels of highly chlorinated polychlorinated biphenyls (PCBs) in the environment, and is a good example of using the combined activity of different microbial communities to detoxify wastes. Results from bench-scale studies at the Environmental Research Laboratory in Athens, Georgia, using sediments collected from the Saginaw River, Ashtabula River, and the Sheboygan Harbor and bay area, suggest that PCBs can be biodegraded under both aerobic and anaerobic conditions. Aerobic bacteria can usually degrade only congeners (members of a family of

River and lake bottom sediments contaminated with chlorinated aromatic compounds can be treated using both anaerobic and aerobic biotreatments.



Thomas Hovland/Grant Heiman Photography Photo

compounds) with one to five chlorine atoms, while anaerobic bacteria can degrade only the more highly chlorinated congeners. This study is examining the effective biodegradation of highly chlorinated PCBs in sediments by sequential anaerobic and aerobic treatments.

Bioremediation and Genetic Engineering

Understanding the genetic and biochemical basis for microbial biodegradation reactions can lead to the innovative construction of microbial gene combinations useful for degrading persistent toxic chemicals.

A gene is essentially the equivalent of a computer program containing information that controls specific biological functions of an organism in relation to its environment. Manipulating this genetic machinery can result in an organism that is better able to degrade a chemical under specific environmental conditions.

Genetic engineering techniques can accomplish this manipulation. This involves identifying and collecting specific strands of DNA from one or more existing microbial species, splicing them together, and inserting the recombinant DNA into another strain. In this way, additional survival and metabolic capabilities can be added to the recipient strain, greatly enhancing its efficiency to degrade target hazardous compounds in a wide range of environmental conditions. To be effective, the recombinant DNA must be maintained in the bacterium and be passed on to subsequent generations.

For bioremediation purposes, genetically engineered microorganisms (GEMs) are still in the re-



Will & Deni McIntyre/Photo Researchers, Inc. Image

search and development stages. The use of such organisms is regulated by the Toxic Substances Control Act, and all genetically altered microbes undergo rigorous safety reviews to evaluate any possible risk to human health or the environment before they are approved for use in the field. To date, GEMs have not been used for site cleanup in the United States.

Genetic engineering research is an exciting and useful technology that has significant future potential. Such research by the EPA may provide microorganisms and biodegradation systems that can destroy persistent, previously undegradable hazardous toxic compounds in the environment.

Hazardous Waste Site Characterization

Site characterization identifies any site-specific problems that must be addressed in applying bioremediation cleanup technology. As previously stated, bioremediation is affected by the types, levels, and distribution of contaminants and the physical nature of the treatment site. All such factors influence the selection of treatment constituents and the engineering methodology for their delivery and maintenance. Because

Deoxyribonucleic acid (DNA) is the heredity molecule. DNA is a long, threadlike macromolecule in which purine and pyrimidine bases (red and yellow in image) carry genetic information while sugar and phosphate groups (green) perform structural roles.

Scientists conduct site characterization activities at a cleanup site prior to selecting treatment.



Don Riepe/Peter Arnold, Inc. Photo

of this, site characterization is a critical phase of bioremediation technology.

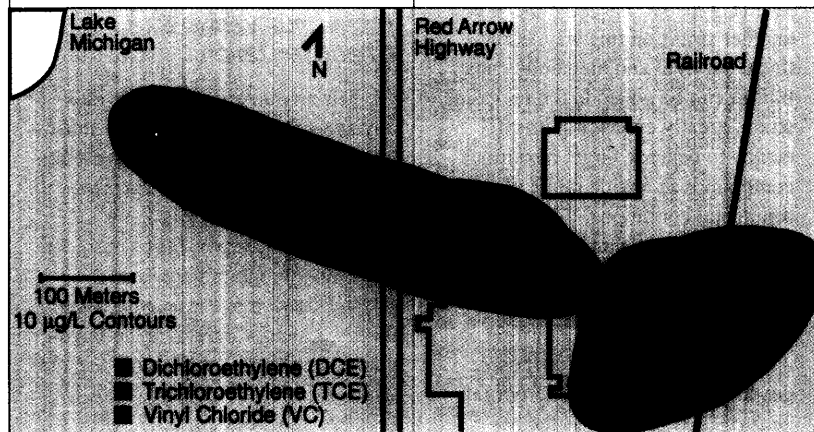
To perform site characterization, scientists and engineers use specialized equipment and surveying, sampling, and soil coring techniques to ascertain all pertinent topographical, structural, and geologic features of the site. They also collect numerous samples that are sent to the laboratory for analysis and classification. Data resulting from these activities

are used to compile site characterization reports that present an accurate composite description of the site and any contamination it contains.

Defining site geological conditions and contaminants present are just part of a comprehensive site characterization. In subsurface soil bioremediation, for example, often a controlling factor is the rate at which the treatment constituents can be successfully applied to the contaminated zone. Soil permeability controls the flux of air or remedial fluids into the contaminated area. Soil

composition (for example, clay and organic matter) has a strong influence on both the rate and extent of bioremediation during land treatment. The capacity of geologic materials to hinder the passage of nutrients often complicates the implementation of bioremediation. All such factors must be considered in the selection of bioremediation technologies. Sometimes the site characterization shows physical or chemical barriers that would prohibit successful bioremediation.

Contour map of a chlorinated aliphatic compound plume found at a National Priority List industrial site in St. Joseph, Michigan.

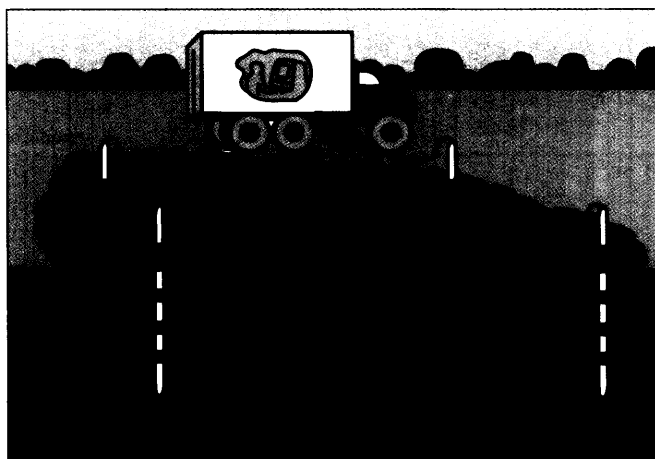


An important aspect of EPA's research in this area involves developing innovative site characterization methods and systems featuring improved reliability, efficiency, and cost-effectiveness. For example, the only approach now available for taking a core sample from the subsurface is to use a hollow stem auger, extract the cores, and determine the quantity of contamination by analytical chemistry techniques. Soil coring is very expensive and carries the risk of spreading the contamination at a site. In collaboration with the U.S. Army Corps of Engineers, ORD has made recent progress toward replacing the hollow stem auger procedure for some applications with a cone penetrometer using fiber optic spectroscopy and on-board computer interpretation for locating and analyzing subsurface waste materials.

ORD is also applying and evaluating a mobile, hydraulically driven soil gas and ground water probe to measure concentrations of hydrocarbons, oxygen, and carbon dioxide. Coupled with established analytical technology, such as field gas chromatographs and infrared cells, this probe will be useful for monitoring and optimizing bioremediation treatment for sites selected for *in situ* bioremediation. These tools will directly improve site characterization by providing efficient and affordable techniques useful for three-dimensionally mapping the distribution of contaminants and measuring the rate of remediation.

Treatment Design and Implementation

Site treatment includes engineering design and field implementa-

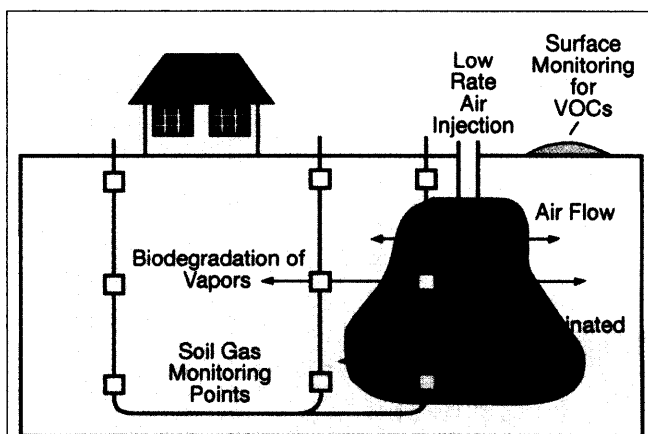


tation of bioremediation applications to physically carry out site cleanup. The techniques and equipment configurations for the remediation of any hazardous waste site are initially selected in response to the various factors identified in the site characterization. Treatment may be required anywhere from the air above the site to the deep subsurface, with treatment *in situ* and/or in above ground systems.

In situ Treatment Techniques

In situ bioremediation techniques are designed to treat the contaminated media in place. Such treatment for soil might include installing irrigation or sprinkler systems to deliver liquid nutrient mixtures directly to the contaminated region to stimulate microorganism growth. If the contaminant is present in the top twelve inches of soil, treatment may also include tilling to aerate the soil. Contamination beneath the surface, where oxygen may be limited, can be treated by installing a series of venting or air injection wells to force air through the soil at low pressure to add oxygen (bioventing). Because bioventing equipment and wells are

A cost-effective Cone Penetrometer with fiber optics and on-board computer will replace hollow stem auger soil coring techniques for some applications.

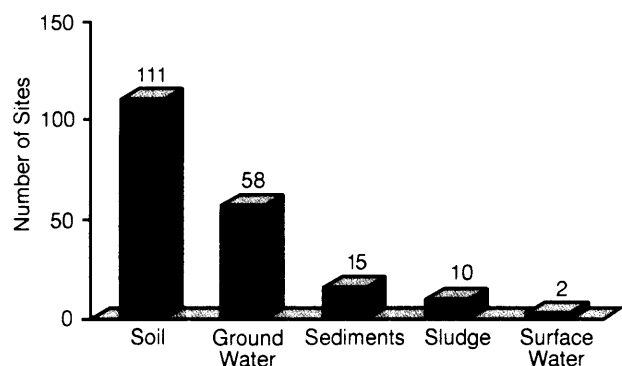


Bioventing introduces oxygen to contaminated soil to stimulate aerobic biodegradation.

Breakdown of the various types of media being treated using bioremediation at more than 150 sites under federal or state regulatory authority.

relatively nonintrusive, this technology is especially valuable for treating contaminated soils in areas where buildings and underground utilities cannot be disturbed. In other cases, injection wells may be used to introduce nutrients and additional oxygen supply (such as hydrogen peroxide) directly to contaminated areas of underground water supplies.

ORD's Risk Reduction Engineering Laboratory (RREL) in Cincinnati, Ohio, is presently conducting two *in situ* bioventing research projects in collaboration with the United States Air Force. The first involves a field bioremediation study at Hill Air Force Base near Salt Lake City, Utah, treating a



site with jet fuel-contaminated soil. The second bioventing project involves remediating a jet fuel spill at Eielson Air Force Base near Fairbanks, Alaska. These bioventing studies are generating valuable pilot-scale performance data and operational experience for a technology that can provide an economic, non-intrusive means of *in situ* cleanup of contaminated soils.

Above Ground Treatment Techniques

EPA scientists and engineers have also developed several above ground bioremediation techniques to treat hazardous wastes on site. These techniques generally use confined areas such as lined treatment beds or enclosed vessels known as bioreactors or biofilters. Bioreactors and biofilters are engineered systems that perform the various stages of the treatment process, such as mixing, nutrient addition, and separation of water and solids. Microorganisms are either freely dispersed or attached as a film to a stationary or moveable surface within the reactor or filter. Treatment in reactors may consist of single or multiple stages. The particular design of a system depends primarily on the characteristics of the site, the type of material to be treated, and the desired treatment results.

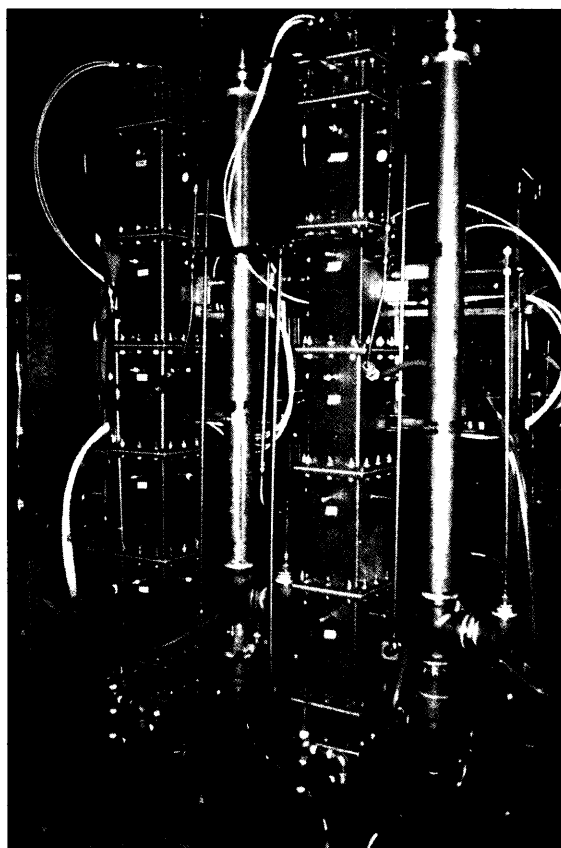
Contaminated materials are placed into or fed through these systems for treatment. Above ground systems provide substantial control of conditions that influence the level of microbial growth and metabolism, such as pH, temperature, oxygen levels, and nutrient concentrations. They also allow for maximum contact between the toxic substance and the microorganisms.

ORD researchers have recently developed an innovative biofilter to control volatile (capable of changing from liquid into gas) organic compound (VOC) emissions from hazardous wastes and contaminated liquid and soil media. The highly efficient system produces complete biodegradation of the VOCs entering the biofilter in as little as two to six minutes under aerobic conditions.

Anaerobic expanded-bed reactors, using granular activated carbon (GAC) as the microorganism support medium, have been developed for efficient removal of chlorinated and non-chlorinated hydrocarbons and other representative hazardous organics in contaminated aqueous wastes and leachates.

Monitoring and Performance Assessment

Built into the site remediation plan is a system for continually monitoring and assessing the progress of the bioremediation treatment and any residual impact to human health or the environment. Research is underway to develop techniques that will streamline the process and reduce costs by providing the most information with the least effort. Such systems might include strategically placed moni-



ORD has developed a highly efficient Air Biofilter system that produces complete biodegradation of VOCs under aerobic conditions.

toring wells, various types of monitoring equipment, and numerous sampling points. Scientists collect and analyze data obtained from these sources to continually assess the biodegradation of the target compounds.



Site personnel take samples at monitoring points to measure bioremediation progress.

Research is designed to improve measures of biodegradation activity. Based on data obtained from monitoring activities, site remediation personnel can make any necessary adjustments to the treatment process, such as increasing the volume of nutrient supplements, to maintain optimal performance.

Bioremediation Used with Other Technologies

Many hazardous waste sites contain complex mixtures of persistent organic and inorganic contaminants that can be cleaned up only by a combination of treatment techniques. For example, highly chlorinated wastes can be effectively treated using chemical methods to dechlorinate the compounds followed by bioremediation to complete detoxification. ORD researchers are developing methods to combine various physical, chemical, and biological treatment technologies, and comparing the effectiveness of the various combinations.

ORD Bioremediation Program

The need to clean up hazardous waste sites as a national priority has accelerated the development of bioremediation technologies. ORD has developed an integrated Bioremediation Program to advance the understanding, development, and application of bioremediation. The program has been designed to strike a balance between basic research activities leading to a fundamental understanding of biodegradation processes and engineering activities leading to practical environmental cleanup applications.

Research, development, and field evaluations are implemented through the efforts of a multi-disciplinary staff of Agency scientists and engineers and are coordinated and directed by the Biosystems Technology Development Steering Committee. This committee, and the broader group of scientists and engineers it represents, constitutes a unique resource in science, innovation,

ORD has developed an integrated bioremediation research program to advance the technology.



SIU/Peter Arnold, Inc. Photo

creativity, and responsiveness to environmental cleanup needs. The program is supplemented by extramural research carried out in concert with other federal agencies, states, contractors, and academic institutions under EPA-funded cooperative agreements, contracts, and interagency agreements.

Bioremediation Program Objectives

The overall goals of ORD's Bioremediation Research Program include the following:

- Identify and characterize biodegradation processes that may be used in the treatment of contaminated surface waters, ground water, sediments, surface and subsurface soils, and gases
- Define, evaluate, optimize, and demonstrate engineering systems necessary for application of the technology to detoxify pollutants *in situ*, on-site, or at centralized treatment facilities
- Develop process-based mathematical models to evaluate potential treatment scenarios and provide a basis for tailoring bioremediation actions to variable chemical contaminants and environmental factors
- Provide protocols and technical assistance to site cleanup managers in selecting appropriate bioremediation technologies for different EPA, regional, state, and local programs
- Formulate a waste site-directed planning framework to characterize sites as suitable for bioremediation
- Develop feedback mechanisms for integrating field information

The Bioremediation Research Program draws on scientists and engineers from the following ORD laboratories and organizations:

- *Environmental Research Laboratory – Athens, GA*
- *Environmental Research Laboratory – Gulf Breeze, FL*
- *Health Effects Research Laboratory – Research Triangle Park, NC*
- *Risk Reduction Engineering Laboratory – Cincinnati, OH*
- *R.S. Kerr Environmental Research Laboratory – Ada, OK*
- *Center for Environmental Research Information – Cincinnati, OH*

from ongoing hazardous waste site cleanup efforts into the research and development process

- Transfer research and technical information to the user community through investigators' meetings, bioremediation workshops, comprehensive resource documents, and a national database on bioremediation field applications

Additional Program Components

Additional components of EPA's Bioremediation Program are the Bioremediation Action Committee and Bioremediation Field Initiative.

The Bioremediation Action Committee (BAC)

ORD chairs and oversees the Bioremediation Action Committee, a working affiliation of experts from government, industry, academia, and the public dedicated to expanding the use of bioremediation in the treat-

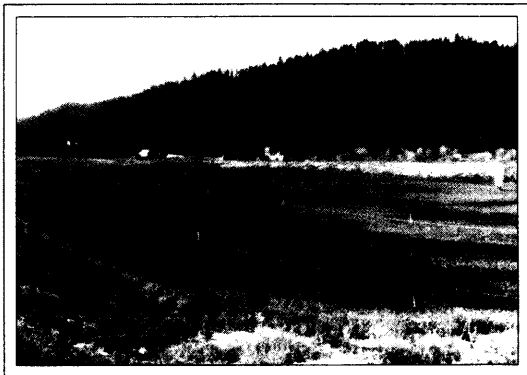
ORD provides technical assistance to EPA regional offices and individual state regulatory agencies overseeing bioremediation projects or considering the use of bioremediation. Technical Support Centers in Ada, Oklahoma, and Cincinnati, Ohio, provide assistance with site characterization, treatability study design, and interpretation of data.

The Bioremediation Field Initiative is Evaluating Three Different Bioremediation Technologies in Progress at a Superfund Cleanup Site in Libby, Montana

Soil and ground water at the Champion International Superfund Site in Libby, Montana, are contaminated by creosote, PAHs, and PCP as a result of past wood treatment operations. The total estimated soil volume requiring treatment today is 45,000 cubic yards (uncompacted), while the plume of contaminated ground water from one of two aquifers at the site extends more than one mile.

Three different bioremediation technologies have been initiated to clean the site, including biological treatment of soils in a prepared-bed land treatment unit (LTU), oil/water separation of ground water followed by biological treatment in a fixed-film bioreactor, and in situ biotreatment of the aquifer.

Under the Bioremediation Field Initiative, ORD scientists from EPA's R. S. Kerr Environmental Research Laboratory in Ada, Oklahoma, provide technical support for remedial activities at the Libby Site. This includes technical direction and oversight for planning, data interpretation, and reporting results from performance evaluation studies being conducted for each of the bioremediation processes in operation.



ment, control, and prevention of environmental contamination. The BAC serves as an important forum for sharing information and collaborative actions across diverse organizations to advance the science and practical field application of bioremediation.

BAC activities include promoting the increased acceptance and use of bioremediation, developing standard protocols for testing bioremediation products and techniques, and coordinating the incorporation of bioremediation in oil and hazardous substance contingency response plans across the United States. The committee also identifies priority research needs, investigates pollution prevention applications of bioremediation, promotes education curricula to adequately prepare scientists and engineers for the field, and facilitates information exchange between EPA and other interested parties on developments and issues regarding federal regulations affecting bioremediation.

Bioremediation Field Initiative

The Bioremediation Field Initiative is a cooperative effort by ORD, EPA's Office of Solid Waste and Emergency Response (OSWER), regional offices, other federal agencies, state agencies, industry, and several universities. The focus of this program is to expand the nation's field experience in bioremediation techniques. Objectives are to more fully assess and document the performance of full-scale field applications and regularly provide information on treatability studies, design, operation, and costs of ongoing bioremediation projects. Another important objective is providing technical assistance to site managers. The initiative is currently tracking

bioremediation activities at more than 150 Superfund, RCRA, and Underground Storage Tank sites.

Conclusion

The growing volume of persistent, difficult-to-treat waste materials accumulating in the environment over the past several decades has contributed to a national pollution problem that affects virtually every community. The United States Environmental Protection Agency is fully committed to reducing, eliminating, or preventing waste materials that threaten human health and the environment. In support of this commitment, the Office of Research and Development will continue its efforts to conduct critical research on cost-effective methods for degrading such materials at their source and in the environment.

Because bioremediation can be an effective, non-disruptive, and cost-efficient means of reducing or eliminating many toxic materials, it should have an ever-increasing role in the successful remediation of hazardous waste and chemical spill sites around the world. The United States is already the world leader in the field implementation of bioremediation technologies. Through its own concerted efforts and activities in collaboration with other programs, ORD will continue to lead in the development, application, and assessment of the technology.

Collaborative Programs

Other programs contributing to the research and development of bioremediation technologies include the following:

Superfund Innovative Technology Evaluation Program

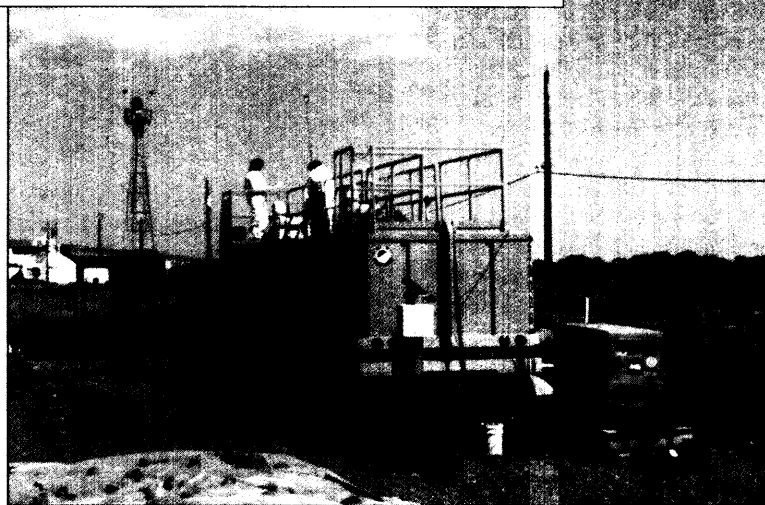
EPA's Superfund Innovative Technology Evaluation (SITE) Program supports the development of innovative treatment technologies for hazardous waste remediation and monitoring/measurement technologies for evaluating the nature and extent of waste site contamination. These include bioremediation technologies. Under the SITE Program, EPA enters into cooperative agreements with technology developers who refine their innovative technologies at the bench-, pilot-, or field-scale and may demonstrate them, with support from EPA, at hazardous waste sites. The SITE Program collects and publishes engineering, performance, and cost data to aid in future decision-making for hazardous waste site remediation. The program is administered by ORD and supported by the Office of Solid Waste and Emergency Response.

Hazardous Substance Research Centers Program

The five cooperative, multi-university Hazardous Substance Research Centers (HSRC) form an integrated national program of basic and applied research, technology transfer, and training. The centers focus on managing hazardous substances and promoting long-term exploratory research to find innovative ways to remediate them. Numerous projects are currently underway exploring bioremediation of contaminated soils and ground water.

Federal Technology Transfer Act Program

The Federal Technology Transfer Act (FTTA) Program supports cooperative research and development agreements between federal laboratories, industry, and academic institutions to develop and commercialize innovative technologies. Such agreements have resulted in the development and field application of several bioremediation technologies and expanded the database of chemicals known to be biodegradable.



One of the pilot-scale technologies evaluated by EPA under the Superfund Innovative Technology Evaluation Program utilizes anaerobic bacterial consortia in a bioreactor (bottom photo) to treat soils contaminated with the pesticide dinoseb (encircled yellow material in soil-top photo). The technology can reduce this pollutant to less than the detection limit in soils and has also been shown to be effective for biodegrading trinitrotoluene (TNT).

EPA Publications

The following reference materials provide more detailed information about the subjects discussed in this document. Copies of these references may be requested at no charge (while supplies are available) from the EPA's Center for Environmental Research Information (CERI). Once the CERI inventory is exhausted, clients will be directed to the National Technical Information Service (NTIS) where documents may be purchased.

Alaskan Oil Spill Bioremediation Project, EPA/600/8-89/073.

Bioremediation Field Initiative Fact Sheets, EPA/540/F-92/012A-J.

Bioremediation in the Field (quarterly newsletter—latest issue August 1993, EPA/540/N-93/002).

Bioremediation of Hazardous Wastes (1990), EPA/600/9-90/041.

Bioremediation of Hazardous Wastes (1991), EPA/600/9-91/036.

Bioremediation of Hazardous Wastes (1992), EPA/600/R-92/126.

Guide for Conducting Treatability Studies Under CERCLA: Aerobic Biodegradation Remedy Screening—Interim Guidance, EPA/540/2-91/013A.

Microbial Decomposition of Chlorinated Aromatic Compounds, EPA/600/2-86/090.

The Federal Technology Transfer Act—Opportunities for Cooperative Biosystems Research and Development with the U.S. EPA, CERI-90-114.

Understanding Bioremediation—A Guidebook for Citizens, EPA/540/2-91/002.

Bioremediation Resource Guide, EPA/542/B-93/004*

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