



Concepts and Technologies for Bioremediation in Confined Disposal Facilities

PURPOSE: The Dredging Operations and Environmental Research (DOER) Program is leveraging with U.S. Army Corps of Engineers (USACE) Districts and the U.S. Environmental Protection Agency to develop low-cost bioremediation technologies for contaminated dredged material in confined disposal facilities (CDFs). This research is aimed at transforming CDFs, diked structures designed to retain dredged material solids (Headquarters, USACE, 1987), from disposal to treatment facilities. Benefits will include operation of CDFs as reclamation facilities, conversion of contaminated dredged material to a soil-like material for beneficial use, and recovery of CDF storage capacity, thereby avoiding the costs of new CDFs. This technical note has two purposes: to assist USACE District staff in their application of bioremediation techniques to organic contaminants in CDFs, and to identify research needed to better understand and optimize contaminant biodegradation in CDFs. Design concepts and bioremediation technologies that show promise for practical application to recalcitrant organic contaminants in dredged material are described. The technologies reviewed include composting, landfarming, and land treatment. Information provided includes description of technologies, applicability, limitations, costs, and the science on which bioremediation technologies are based.

BACKGROUND: Contaminated sediments in many industrial and urbanized harbors and waterways contribute to environmental degradation and hinder the USACE in the performance of its navigation mission. In the course of accomplishing its mission of maintaining and improving navigation in waters of the United States, the USACE annually handles about 300 million cubic meters of dredged material. Five to ten percent of this material is not suitable for unrestricted open-water disposal. Recalcitrant organic compounds, such as polychlorinated dibenzo-*p*-dioxins/furans (PCDDs/Fs), polychlorinated biphenyls (PCBs), and polycyclic aromatic hydrocarbons (PAHs), have drawn attention as threats to ecological and human health. The presence of these contaminants in sediments to be dredged often results in significant project delays and management cost increases. Currently, contaminated dredged material unsuitable for unrestricted open-water disposal may be confined in CDFs and aquatic disposal sites or treated.

Many CDFs are nearing or exceeding design capacity. Acquisition of land for new CDFs is difficult because choice land areas located near dredging projects are already in use and undeveloped lands near dredging projects are frequently wetlands whose ecological functions make them too valuable for use as CDFs. A solution to this problem is to extend the service life of existing CDFs as much as possible (Montgomery et al. 1978).

CDF closure studies conducted by the U.S. Army Engineer District, Detroit, have shown that PAH concentrations are lower in the upper unsaturated zone than in the saturated zone (Myers and Bowman 1999). These results are consistent with the top-down, self-cleaning hypothesis (Myers 1996) involving intrinsic biodegradation and clearly show that the top meter of material can be substantially cleaner than the deeper material in CDFs. Bioremediation, therefore, may offer an

effective and affordable means of converting CDFs from containment facilities to dredged material reclamation facilities. However, at present it is difficult to determine with certainty if bioremediation is a viable alternative for treatment of contaminated dredged materials.

OVERVIEW OF CONCEPTS AND TECHNOLOGIES: Bioremediation techniques stimulate microorganisms to grow and use contaminants as food and energy sources by creating a favorable environment for the microorganisms. Generally, this means providing some combination of oxygen, nutrients, and moisture, and controlling temperature and pH. Sometimes, microorganisms adapted for degradation of specific contaminants are added to enhance the process.

Biological processes typically are implemented at lower cost than physical and chemical treatment processes. Contaminants can be broken down (destroyed) by biodegradation through a process called catabolism. However, biological processes typically require more time than physical and chemical treatment processes, and it is sometimes difficult to determine whether contaminants have been actually destroyed by biological processes. When a contaminant has been catabolized to the stable end products carbon dioxide and water, the contaminant is said to have been mineralized.

Although not all organic compounds are amenable to biodegradation, bioremediation techniques have been used successfully to remediate soils, sludges, and groundwater contaminated by petroleum hydrocarbons, chlorinated solvents, wood preservatives, explosives, and other organic chemicals (Flathman, Jerger, and Exner 1994; Norris et al. 1994; Bajpai and Zappi 1997). The rate at which contaminants are biologically degraded is influenced by the specific contaminants present and their concentrations, oxygen supply, moisture, temperature, pH, nutrient supply, and the microbial consortia present.

Bioremediation may be carried out in situ or ex situ. In situ treatment proceeds without excavation and transportation, resulting in significant cost savings. However, in situ treatment generally requires longer time periods and provides less uniformity of treatment. Available in situ biological treatment technologies include bioventing, land treatment, phytoremediation, and natural attenuation. Ex situ treatment generally requires shorter time periods than in situ treatment and provides more uniformity of treatment because of the ability to homogenize, screen, continuously mix, and monitor. However, ex situ treatment requires excavation, leading to increased costs and engineering for equipment. Available ex situ biological treatment technologies include composting, landfarming, and slurry phase biological treatment.

Treatability studies typically are conducted to determine the effectiveness of bioremediation in a given situation. The extent of the study can vary depending on the nature of the contaminants and the characteristics of the site. For sites contaminated with common petroleum hydrocarbons (e.g., gasoline and/or other readily biodegradable compounds), it is usually sufficient to examine representative samples for the presence and level of an indigenous population of microbes, nutrient levels, presence of microbial toxicants, and soil characteristics such as pH, porosity, and moisture. Treatability studies on dredged material contaminated with hydrophobic organics, on the other hand, require bench-, pilot-, and demonstration-scale tests to identify cost-effective designs and operating procedures. In addition, statistical characterization techniques are needed to represent “before” and “after” situations for heterogeneous materials, such as dredged material, to verify biological treatment effectiveness.

CANDIDATE TECHNOLOGIES FOR BIOREMEDIATION IN CDFs: In this section, selected bioremediation technologies with potential for cost-effective application to CDFs are briefly described, compared, and contrasted. The scientific basis for application of bioremediation technologies to recalcitrant organic compounds, such as PAHs, PCBs, and PCDDs/Fs, is presented in Appendix I. The technologies selected for evaluation are composting, landfarming, and land treatment.

Composting. Composting is a controlled biological process by which organic contaminants are converted by microorganisms to innocuous, stabilized by-products. Typically, thermophilic conditions (54 to 65 °C) must be maintained to properly compost soil contaminated with hazardous organic contaminants. The increased temperatures result from heat produced by microorganisms during the degradation of organic material. In most cases, soil composting uses indigenous microorganisms and the addition of readily available and biodegradable animal or vegetable wastes (e.g., sewage sludge). Soils are excavated and mixed with organic amendments and bulking agents, such as wood chips, to enhance the porosity of the mixture to be composted. Maximum degradation efficiency is achieved through maintaining oxygenation (e.g., daily windrow turning), irrigating as necessary, and closely monitoring moisture content and temperature.

Three process designs are used in composting: mechanically agitated in-vessel composting (compost is placed in a reactor vessel where it is mixed and aerated), windrow composting (compost is placed in long piles known as windrows and periodically mixed with mobile equipment), and aerated static pile composting (compost is formed into piles and aerated with blowers or vacuum pumps). Windrow composting (Figure 1) is usually considered to be the most cost-effective composting alternative. It also may have the highest fugitive gas emissions.



Figure 1. Turning of compost windrows at the Jones Island CDF, Milwaukee, WI (from Myers and Bowman 1999)

Biopile treatment (Figure 2) is another full-scale composting technology in which excavated soils are mixed with soil amendments and placed on a treatment area that includes some form of aeration (von Fahnestock et al. 1998). Biopile composting differs from windrow composting in that the pile is not mixed once constructed. Soil biopiles commonly have an air distribution system buried in the treatment area to pass air through the soil either by vacuum or by positive pressure. The soil piles in this case can be up to 6 m (20 ft) high (generally not recommended, 2-3 m maximum). Biopiles may be covered with plastic to control runoff, evaporation, and volatilization and to promote solar heating. Vendors have developed proprietary nutrient and additive formulations and methods for incorporating the formulations into the soil to stimulate biodegradation. The formulations are usually modified for site-specific conditions. Moisture, heat, nutrients, oxygen, and pH can be controlled to enhance biodegradation.

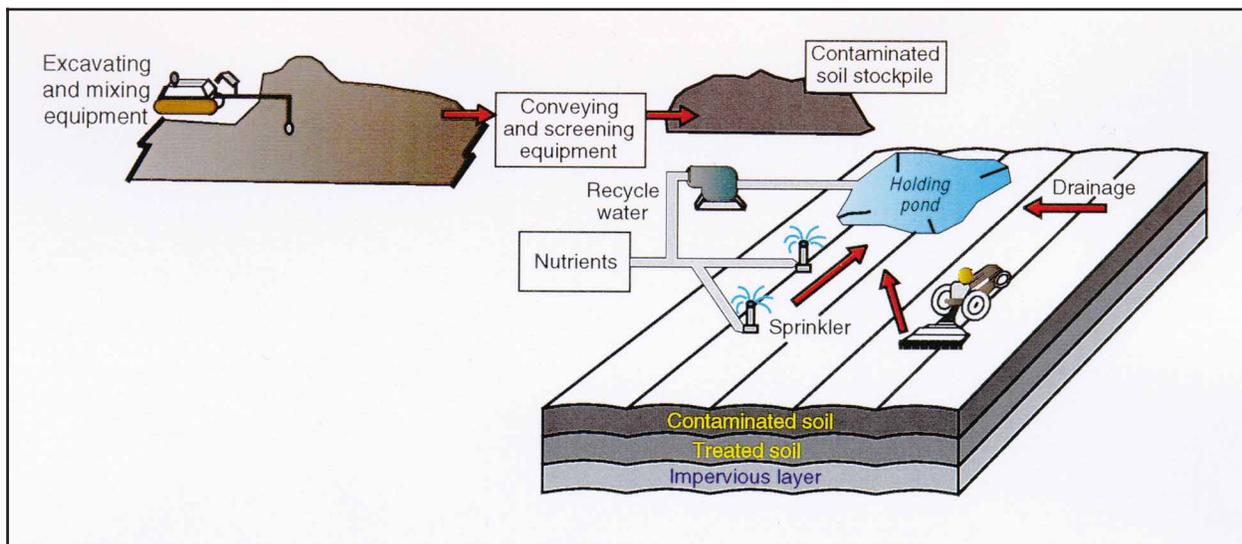


Figure 2. Schematic of typical biopile treatment system (from Federal Remediation Technologies Roundtable: Remediation Technologies Screening Matrix and Reference Guide Version 3.0, <http://www.frtr.gov>)

Landfarming. Landfarming (Figure 3) is a full-scale bioremediation technology that usually incorporates liners and other methods to control leaching of contaminants and requires excavation and placement of contaminated soils. Contaminated media are applied into lined beds and periodically turned over or tilled to aerate. Soil conditions are often controlled to optimize the rate of contaminant degradation. Conditions normally controlled include the following:

- Moisture content usually by irrigation or spraying.
- Aeration by tilling the soil at a predetermined frequency.
- pH (buffered near neutral pH by adding crushed limestone or agricultural lime).
- Other amendments (e.g., soil bulking agents, nutrients, etc.).

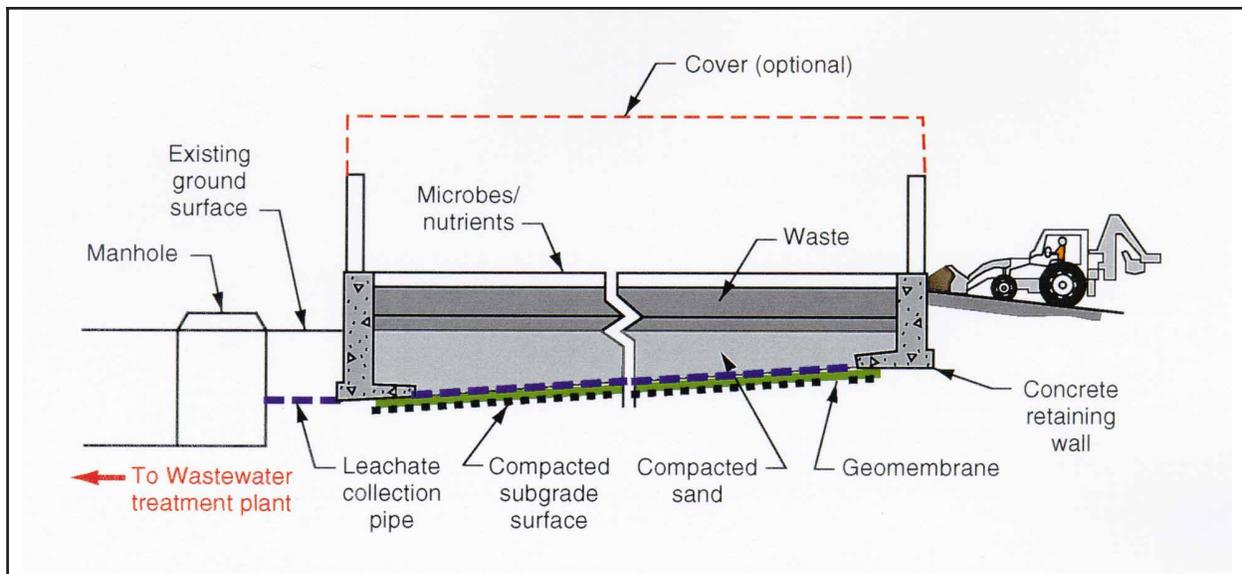


Figure 3. Schematic of typical landfarming treatment unit (from Federal Remediation Technologies Roundtable: Remediation Technologies Screening Matrix and Reference Guide Version 3.0, <http://www.frtr.gov>)

Contaminated media are usually treated in lifts that are up to 0.46 m (18 in.) thick. When the desired level of treatment is achieved, the lift is removed and a new lift is constructed. It may be desirable to remove only the top of the remediated lift, then construct the new lift by adding more contaminated media to the remaining material and mixing. This serves to inoculate the freshly added material with an actively degrading microbial culture, which can reduce treatment times. Landfarming is a medium- to long-term technology.

Land Treatment. Land treatment is a full-scale bioremediation technology in which contaminated soils, sediments, or sludges are periodically turned over (i.e., tilled) and allowed to interact with the soil and climate at the site. One advantage of land treatment is that conventional farming equipment (Figure 4) is usually the only equipment required.

The contaminants, soil, climate, and biological activity interact dynamically as a system to degrade, transform, and immobilize contaminants. Soil conditions are often controlled to optimize the rate of contaminant biodegradation. Conditions normally controlled include those previously listed for landfarming. A land treatment unit must be managed properly to prevent both onsite and offsite problems with groundwater, surface water, air, or food chain contamination. Adequate monitoring and environmental safeguards are required.

COMPARISON OF TECHNOLOGIES: Logistically, all the technologies discussed in this technical note are fully developed. Equipment and procedures are well known for some applications. Table 1 summarizes attributes of the four technologies described.



Figure 4. Typical land treatment equipment

Table 1 Attributes of Selected Candidate Bioremediation Technologies for Dredged Material*				
Attribute	Windrow Composting	Landfarming	Biopile Composting	Land Treatment
Applications	Soil, lagoon sediment; explosives, PAHs	Diesel fuel, fuel oil, PCB, pesticides	Fuels, solvents	Fuels and oils, wood preserving waste
Development status	Full	Full	Full	Full
Site preparation and equipment	Excavation req'd; special mixing equipment	Excavation req'd; simple earthmoving equipment	Excavation req'd; simple earthmoving equipment	Conventional farming equipment
Limitations	Bulking agents increase vol. and may req removal for end use	Permanent type structure req'd	Static processes may treat less uniformly than those with mixing	Erosion/runoff potential
Cost, \$ per cu m (\$ per cu yd)	\$248 (\$190)	<\$98 (\$75)	\$35 (\$27);* \$52-131 (\$40-100)**	\$33-65 (\$25-50)
<p>* Federal Remediation Technologies Roundtable; Remediation Technologies Screening Matrix and Reference Guide Version 3.0, http://www.frtr.gov/matrix2/top_page.html. ** Data from von Fahnestock et al. (1998).</p>				

Composting, landfarming, and biopiles require excavation and placement in a pile, row, or containment facility. These technologies offer greater control, at greater cost. Composting involves aggressive use of bulking agents and requires addition of readily biodegradable organic matter. These amendments increase volume and may be problematic for end-use applications, requiring a separations step before use as building materials or fill. In addition, questions also may be raised as to the extent of destruction, versus sequestering of contaminants by the substantial organic matter added.

Scale and permanence of operation are also factors affecting costs. Table 2 illustrates the impact of these two factors on biopile composting costs. Cost estimates for biopiles range from \$54/cu m (\$41/cu yd) to almost \$260/cu m (\$200/cu yd) depending on facility scale and permanence.

Table 2 Cost Analysis for Biopile Treatment*		
Total Biopile Volume cu m (cu yd)	Cost at Temporary Facility \$/cu m (\$/cu yd)	Cost at Permanent Facility \$/cu m (\$/cu yd)
382 (500)	135 (103)	257 (197)
3,823 (5,000)	54 (41)	65 (50)
30,580 (40,000)	—	54 (41)
* Data from von Fahnestock et al. (1998); assumes no transportation costs, free utilities, a 4-month treatment cycle, and permitting completed.		

Clearly, scale matters, and costs can be lowered by establishing a permanent capability to treat large volumes. CDFs hold large volumes of dredged material and range in size from about 4 hectares (10 acres) to 810 hectares (2,100 acres). A 4-hectare (10-acre) section treated, for example, to a depth of 1.8 m (6 ft) represents approximately 76,460 cu m (100,000 cu yd) of volume (before addition of any bulking agents). Beyond size, institutional support at the site will play a role in costs. For example, the cost estimate listed in Table 2 for a temporary 3,823-cu-m (5,000-cu-yd) biopile facility included about \$39,000 for labor costs, including weekly inspections, record keeping, grounds keeping, administrative support, respiration tests, and soil sampling.

Lower cost than indicated in Table 1 for composting and biopile technologies may be realized at CDFs that do not require liners and leachate collection systems. There also may be opportunities for cost reduction at those sites where personnel are on hand for other work, and tasks can be integrated. Remote and unattended sites to which personnel must travel for all maintenance and monitoring will have correspondingly higher costs.

If construction of a new CDF is an alternative, then the cost of remediation in an existing CDF can be weighed against new construction costs. New CDF construction costs in the range of \$20-33/cu m (\$15-25/cu yd) effectively put a “cap” on remediation expenditures, unless other factors justify a higher expenditure. For these reasons, end use will have a significant impact on the economic viability and choice of remediation methods. Beneficial uses, e.g., building materials, construction fill, and manufactured soil, may provide a payback that will offset the cost of treatment. Open-water

disposal of treated dredged material may provide a “no payback” option. However, regulatory and public acceptance issues limit this option.

RESEARCH AND DEVELOPMENT PRIORITIES: Two broad categories encompass research and development needs for advancing CDF technology beyond storage and containment to a treatment and, desirably, a reuse paradigm that would allow recovery of storage capacity as follows:

- Field practice needs: guidelines for altering construction and/or management practices to accelerate aerobic bioremediation of dredged material; personnel, time, and equipment needs; cost estimates.
- Process design needs: rates and extent of biodegradation; factors affecting and optimum conditions for biodegradation; relative effects of volatilization and photolysis; development of aerobic zone (transport of oxygen in dredged material) and related concerns, e.g., drying.

These research needs must be addressed simultaneously due to the immediate need to recover storage capacity in CDFs and DOER program requirements (approximately 3-year work unit efforts). A long, deliberate research program involving extensive laboratory studies that are eventually followed by pilot- and demonstration-scale tests will not fill the immediate need for new contaminated dredged material management technologies nor is such an approach consistent with DOER program requirements. On the other hand, a few field demonstrations in the absence of bench-scale treatability studies are not likely to define the critical correlations needed to eventually develop design guidance. Thus, simultaneous and parallel efforts in the field and the laboratory are recommended.

The field efforts should focus on obtaining detailed performance and cost data on bioremediation technologies. Equipment productivity, overall costs (equipment, monitoring, and personnel), and descriptions of lessons learned (i.e., mistakes) should be documented in each field effort. Dredged material in CDFs is highly heterogeneous with respect to chemical and physical properties. Research on effective and economical statistical characterization techniques is needed to describe treatment performance with confidence and accuracy. Inexpensive assay techniques, such as described by McFarland, McCant, and Inouye (1998) for dioxin, should be used to the maximum extent possible to reduce the cost of field monitoring. In addition, volatilization should be monitored in pilot- and demonstration-scale tests to show that the contaminants being biodegraded are not simply volatilizing.

The major process unknowns include time requirements for treatment (rate and extent of reaction), optimal use of nutrients, bioaugmentation, air, water, and oxygen, and the specifics of anaerobic to aerobic transition in CDFs. The time requirement (rate and extent of reaction) is the most important unknown. Field biodegradation rates for recalcitrant organic compounds in dredged material are unknown, and this information must be obtained to evaluate the technical feasibility of dredged material bioremediation and develop design guidance. There are also questions about the extent to which biodegradation can proceed. Recent research has indicated that chemicals such as PAHs somehow become sequestered such that they are not bioavailable and therefore not biodegradable (Luthy et al. 1999). Rates and extent of reaction and the factors affecting biodegradation rates and the extent to which dredged material can be cleaned by biodegradation should be determined in

bench-scale treatability studies. Bench-scale treatability studies are also needed to determine why the available data show no breakdown of PAHs during composting of dredged material (Myers and Bowman 1999) but PCBs disappear during both composting (Myers and Bowman 1999) and land treatment (Tang and Myers 1998).

SUMMARY: Numerous laboratory studies have shown that recalcitrant organic compounds, such as of PAHs, PCBs, and PCDDs/Fs, can be biodegraded (Appendix I). This literature provides the scientific basis for developing bioremediation technologies for dredged material contaminated with these compounds. Composting and land treatment technologies have the potential for successful and cost-effective degradation of the compounds of concern discussed herein. However, sufficient understanding to assess bioremediation as a viable treatment alternative of contaminated dredged material is lacking. Additional research, especially at the pilot- and demonstration-scale, is needed to better understand process kinetics, practical implementation scenarios, and costs.

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Myers, T. E., and Williford, C. W. (2000). "Concepts and technologies for bioremediation in Confined Disposal Facilities," *DOER Technical Notes Collection* (ERDC TN-DOER-C11), U.S. Army Engineer Research and Development Center, Vicksburg, MS. www.wes.mil/el/dots/doer

REFERENCES

- Abramowicz, D. A., Brennan, M. J., and van Dort, H. M. (1990). "Anaerobic and aerobic biodegradation of endogenous PCBs." *Research and Development Program for the Destruction of PCBs*, Ninth Progress Report, General Electric Corporation, Corporate Research and Development, Schenectady, NY.
- Adriaens, P., and Grbic'-Galic', D. (1994). "Reductive dechlorination of PCDD/F by anaerobic cultures and sediments," *Chemosphere* 29, 2253-2259.
- Adriaens, P., Fu, Q., and Grbic'-Galic', D. (1995). "Bioavailability and transformation of highly chlorinated dibenzo-*p*-dioxins and dibenzofurans in anaerobic soils and sediments," *Environmental Science and Technology* 29, 2252-2261.
- Adriaens, P., Chang, P. R., and Barkovskii, A. L. (1996). "Dechlorination of chlorinated PCDD/F by organic and inorganic electron transfer molecules in reduced environments," *Chemosphere* 32, 433-441.
- Albrecht, I. D., Barkovskii, A. L., and Adriaens, P. (1999). "Production and dechlorination of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin in historically-contaminated estuarine sediments," *Environmental Science and Technology* 33 (5), 737-744.
- Alder, A. C., Haggblom, M. M., Oppenbeimes, S. R., and Young, L. Y. (1993). "Reductive dechlorination of polychlorinated biphenyls in anaerobic sediments," *Environmental Science and Technology* 27, 530-536.
- Bajpai, R. K., and Zappi, M. E. (1997). *Bioremediation of surface and subsurface contamination*. New York Academy of Sciences, New York.
- Barkovskii, A. L., and Adriaens, P. (1995). "Reductive dechlorination of tetrachlorodibenzo-*p*-dioxin partitioned from historically-contaminated sediments to an allochthonous microbial community," *Organohalogen Compounds* 24, 17-21.

- Bedard, D. L., Unterman, R. D., Bopp, L. H., Brennan, M. J., Haberl, M. L., and Johnson, J. (1986). "Rapid assay for screening and characterizing microorganisms for the ability to degrade polychlorinated biphenyls," *Applied Environmental Microbiology* 52, 761-768.
- Beurskens, J. E. M., Toussaint, M., de Wolf, J., van der Steen, J. M. D., Slot, P. C., Commandeur, L. C. M., and Parsons, H. H. (1995). "Dehalogenation of chlorinated dioxins by an anaerobic microbial consortium from sediment," *Environmental Toxicology and Chemistry* 14, 939-943.
- Brown, J. F., Jr., Bedard, D. L., Brennan, M. J., Carnahan, J. C., Feng, H., and Wagner, R. E. (1987a). "Polychlorinated biphenyl dechlorination in aquatic sediments," *Science* 236, 709-712.
- Brown, J. F., Jr., R Wagner, R. E., Feng, H., Bedard, D. L., Brennan, M. J., Carnahan, J. C., and May, J. (1987b). "Environmental dechlorination of PCBs," *Environmental Toxicology and Chemistry* 6, 579-593.
- Bumpus, J. A., Tien, M., Wright, D., and Aust, S. D. (1989). "Oxidation of persistent environmental pollutants by a white rot fungus," *Science* 228, 1434-1436.
- Bunyan, P. J., and Page, J. M. J. (1978). "Polychlorinated biphenyls, the effect of structure on the induction of quail hepatic microsomal enzymes," *Toxicology and Applied Pharmacology* 45, 507-518.
- Bush, B., Simpson, K. W., Shane, L., and Koblinz, R. R. (1985). "PCB congener analysis of water and caddisfly larvae (Insecta: Trichoptera) in the Upper Hudson River by glass capillary chromatography," *Bulletin of Environmental Contamination and Toxicology* 34, 96-105.
- Cerniglia, E. E. (1984). "Microbial metabolism of polycyclic aromatic hydrocarbons," *Advances in Applied Microbiology* 30, 31-71.
- Choudhary, G., Keith, L. H., and Rappe, C. (1983). *Chlorinated dioxins and dibenzofurans in the total environment*. Butterworth Publishers, Boston, MA.
- Christian, B. J., Pugh, L. B., and Clarke, B. H. (1995). "Aromatic hydrocarbon degradation in hydrogen peroxide- and nitrate-amended microcosms." *Bioremediation of recalcitrant organics*. R. E. Hinchee and R. E. Hoeppe, ed., Battelle Press, Columbus, OH.
- Clarke, A. N., Megehee, M. M., Lowe, D. L., and Clarke, J. H. (1994). "A review of polychlorinated dibenzofuran and polychlorinated dibenzodioxins in sediments in the United States and international waterways," *Hazardous Waste and Hazardous Materials* 11, 253-275.
- Clarke, J. U., McFarland, V. A., and Pierce, B. D. (1989). "Preliminary recommendations for a congener-specific PCB analysis in regulatory evaluation of dredged material," Miscellaneous Paper D-89-2, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.
- Davis, M. W., Glaser, J. A., Evans, J. W., and Lamar, R. T. (1993). "Field evaluation of the lignin-degrading fungus *Phanerochaete sordida* to treat creosote-contaminated soil," *Environmental Science and Technology* 27, 2572-2576.
- Dreyer, G., Konig, J., and Ringpfeil, M. (1995). "Polycyclic aromatic hydrocarbon biodegradation by a mixed bacterial culture." *Bioremediation of recalcitrant organics*. R. E. Hinchee and R. E. Hoeppe, ed., Battelle Press, Columbus, OH.
- Duinker, J. C., and Hillebrand, M. T. J. (1983). "Composition of PCB mixtures in biotic and abiotic marine compartments (Dutch Wadden Sea)," *Bulletin of Environmental Contamination and Toxicology* 31, 25-32.
- Duinker, J. C., Hillebrand, M. T. J., and Boon, J. P. (1983). "Organochlorines in benthic invertebrates and sediments from the Dutch Wadden Seas; Identification of individual PCB components," *Netherlands Journal of Sea Research* 17(1), 19-38.
- Efroymsen, R. A., and Alexander, M. (1991). "Biodegradation by an *Arthrobacter* species of hydrocarbons in micellar nonionic surfactant solutions," *Environmental Science and Technology* 57, 1441-1447.
- Federal Remediation Technologies Roundtable: Remediation Technologies Screening Matrix and Reference Guide Version 3.0. <http://www.frtr.gov>
- Flathman, P. E., Jerger, D. E., and Exner, J. H. (1994). *Bioremediation field experience*. Lewis Publishers, Ann Arbor, MI.

- Hammel, K. E., Kalyanaraman, B., and Kent-Kirk, T. (1986). "Oxidation of polycyclic aromatic hydrocarbons and dibenzo-(p)-dioxins by *Phanerochaete chrysosporium* ligninase," *Journal of Biological Chemistry* 261, 16948-16952.
- Harkness, M. R., McDermott, J. B., Abramowicz, D. A., Salvo, J. J., Flannagan, W. P., Stephens, M. L., Mondello, F. J., May, R. J., Lobos, J. H., Carroll, K. M., Brennan, M. J., Bracco, A. A., Fish, K. M., Warner, G. L., Wilson, P. R., Dietrich, D. K., Lin, D. T., Morgan, C. B., and Gately, W. L. (1993). "In situ stimulation of aerobic PCB biodegradation in Hudson River sediments," *Science* 259, 503-507.
- Headquarters, U.S. Army Corps of Engineers. (1987). "Confined disposal of dredged material," Engineer Manual 1110-2-5027, Washington, DC.
- Jee, V., Beckles, D. M., Ward, C. H., and Hughes, J. B. (1998). "Aerobic slurry reactor treatment of phenanthrene contaminated sediment," *Water Research* 32(4), 1231-1239.
- Jones, W. J., Araujo, R., and Rogers, J. E. (1996). "Bench-scale evaluation of bioremediation for the treatment of sediments from the Ashtabula, Buffalo, Saginaw, and Sheboygan Rivers," EPA 905-R96-012, U.S. Environmental Protection Agency, Great Lakes National Program Office, Chicago, IL.
- Keck, J., Sims, R. C., Cooper, M., Park, K. D., and Symons, B. (1989). "Evidence of co-oxidation of polycyclic nuclear aromatic hydrocarbons in soil," *Water Research* 23(12), 1467-1476.
- Klasson, K. T., Barton, J. W., Evans, B. S., and Reves, M. E. (1996). "Reductive microbial dechlorination of indigenous polychlorinated biphenyls in soil using a sediment-free inoculum," *Biotechnology Progress* 12, 310-315.
- Luthy, R. G., Ghosh, U., Talley, J. W., Gillette, J. S., and Zare, R. N. (1999). "Microscale investigation of bioavailability and biostabilization of PAHs in sediments," presentation at the Milwaukee CDF Composting Demonstration Visitors' Day, 22 September 1999, Milwaukee, WI.
- May, R., Schroder, P., and Sandermann, H., Jr. (1997). "Ex-situ process for treating PAH-contaminated soil with *Phanerochaete chrysosporium*," *Environmental Science and Technology* 31(9), 2626-2633.
- McFarland, V. A., Clarke, J. U., and Gibson, A. B. (1986). "Changing concepts and improved methods for evaluating the importance of PCBs as dredged sediment contaminants," Miscellaneous Paper D-86-5, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.
- McFarland, V. A., McCant, D. D., and Inouye, L. S. (1998). "Guidance for performance of the H4IIE dioxin screening assay," *DOER Technical Notes Collection* (TN-DOER-C1), U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS. www.wes.army.mil/el/dots/doer/
- Mohammed, S. A., Sorensen, D. L., Sims, R. C., and Sims, J. L. (1998). "Pentachlorophenol and phenanthrene biodegradation in creosote contaminated aquifer material," *Chemosphere* 37(1), 103-111.
- Montgomery, R. L., Ford, A. W., Poindexter, M. E., and Bartos, M. J. (1978). "Guidelines for dredged material disposal area reuse management," Technical Report DS-78-12, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.
- Mueller, J. G., Chapman, P. J., and Pritchard, H. P. (1989). "Action of a fluoranthene-utilizing bacterial community on polycyclic aromatic hydrocarbon components of creosote," *Applied Environmental Microbiology* 55(12), 3085-3090.
- Mueller, J. G., Lantz, S. E., Devereus, R., Berg, J. D., and Pritchard, P. H. (1994). "Studies on the microbial ecology of polycyclic aromatic hydrocarbon biodegradation." *Bioremediation of chlorinated and polycyclic aromatic hydrocarbons compounds*. R. E. Hinchee, A. Leeson, L. Semprini, and S. K. Ong, ed., Lewis Publishers, Boca Raton, FL.
- Myers, T. E. (1996). "Natural processes for contaminant treatment and control at dredged material confined disposal facilities," *Environmental Effects of Dredging Technical Notes Collection* (EEDP-02-19), U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS. www.wes.army.mil/el/dots/
- Myers, T. E., and Bowman, D. W. (1999). "Bioremediation of PAH-contaminated dredged material at the Jones Island CDF: Materials, equipment, and initial operations," *DOER Technical Notes Collection* (TN-DOER-C5), U.S. Army Engineer Research and Development Center, Vicksburg, MS. www.wes.army.mil/el/dots/doer/

- Myers, T. E., and Tang, N. H. (1998). "Natural attenuation of dioxins in exposed dredged material." *Hazardous and industrial wastes — Proceedings of the 30th Mid-Atlantic Industrial and Hazardous Waste Conference*. R. P. S. Suri and G. L. Christensen, ed., Battelle Press, Columbus, OH.
- Natarajan, M. R., Wu, W., Nye, J., Wang, H., and Jain, M. K. (1996). "Dechlorination of polychlorinated biphenyl congeners by an anaerobic microbial consortium," *Applied Microbiology and Biotechnology* 43, 673-677.
- Natarajan, M. R., Wu, W., Wang, H., Bhatnagar, L., and Jain, M. K. (1998). "Dechlorination of spiked PCBs in lake sediment by anaerobic microbial granules," *Water Research* 32(10), 3013-3020.
- National Research Council. (1979). *Polychlorinated biphenyls*. National Academy of Sciences, Washington, DC.
- Nies, L., and Vogel, T. M. (1990). "Effects of organic substrates on dechlorination of Aroclor 1242 in anaerobic sediments," *Applied Environmental Microbiology* 56, 2614-2617.
- Norris, R. D., Hinchee, R. E., Brown, R., McCarty, P. L., Semprini, L., Wilson, J. T., Kampbell, D. H., Reinhard, M., Bouwer, E. J., Borden, R. C., Vogel, T. M., Thomas, J. M., and Ward, C. H. (1994). *Handbook of bioremediation*. Lewis Publishers, Ann Arbor, MI.
- Olie, K., Vermeulen, P. L., and Hutzing, O. (1977). "Chlorodibenzo-*p*-dioxins and chlorodibenzofurans are trace compounds of fly ash and flue gas of some municipal incinerators in The Netherlands," *Chemosphere* 8, 455-459.
- Pott, B. M., and Henrysson, T. (1995). "Ex situ bioremediation of polycyclic aromatic hydrocarbons in laboratory systems." *Bioremediation of recalcitrant organics*. R. E. Hinchee and R. E. Hoeppe, ed., Battelle Press, Columbus, OH.
- Quensen, J. F., III, Tiedje, J. M., and Boyd, S. A. (1988). "Reductive dechlorination of polychlorinated biphenyls by anaerobic microorganisms from sediments," *Science* 242, 752-754.
- Quensen, J. F., III, Boyd, S. A., and Tiedje, J. M. (1990). "Dechlorination of four commercial polychlorinated biphenyl mixtures (Aroclors) by anaerobic microorganisms from sediments," *Applied Environmental Microbiology* 56, 2360-2369.
- Rappe, C., Choudhary, G., and Keith, L. H. (1986). *Chlorinated dioxins and dibenzofurans in perspective*. Lewis Publishers, Inc., Chelsea, MI.
- Sims, J. C., and Overcash, M. R. (1983). "Fate of polynuclear aromatic compounds (PNAs) in soil-plant systems," *Residue Reviews* 88, 1-68.
- Sims, R. C., Doucette, W. J., Mclean, J. E., Grenney, W. J., and DuPont, R. R. (1988). "Treatment potential for 56 EPA-listed hazardous chemicals in soil," EPA/600/6-88/001, Robert S. Kerr Environmental Research Laboratory, Ada, OK.
- Tang, N. H., and Myers, T. E. (1998). "Natural attenuation of PCB in dredged sediment." *Hazardous and industrial wastes — Proceedings of the 30th Mid-Atlantic Industrial and Hazardous Waste Conference*. R. P. S. Suri and G. L. Christensen, ed., Battelle Press, Columbus, OH.
- Van Dort, H. M., and Bedard, D. L. (1991). "Reductive ortho- and meta-dechlorination of a polychlorinated biphenyl congener by anaerobic microorganism," *Applied Environmental Microbiology* 57, 1576-1578.
- von Fahnestock, F. M., Wickramanayake, G. B., Kratzke, R. J., and Major, W. R. (1998). *Biopile design, operation, and maintenance handbook for treating hydrocarbon-contaminated soils*. Battelle Press, Columbus, OH.
- Williams, W. A., and May, R. J. (1997). "Low-temperature microbial aerobic degradation of polychlorinated biphenyls in sediment," *Environmental Science and Technology* 31(12), 3491-3496.
- Wilson, S. C., and Jones, K. C. (1993). "Bioremediation of soil contaminated with polynuclear aromatic hydrocarbons (PAHs): A review," *Environmental Pollution* 81, 229-249.
- Wu, Q., Bedard, D. L., and Weigel, J. (1999). "2,6-Dibromobiphenyl primes extensive dechlorination of aroclor 1260 in contaminated sediment at 8-30 °C by stimulating growth of PCB-dehalogenating microorganisms," *Environmental Science and Technology* 33(4), 595-602.

APPENDIX I SCIENTIFIC BASIS FOR BIOREMEDIATION OF RECALCITRANT ORGANIC COMPOUNDS

PAHs. PAHs are a diverse group of organic compounds containing two or more fused benzene rings in various configurations, with or without substituent groups. These compounds are neutral, nonpolar, and hydrophobic; each PAH exhibits a unique set of physical and chemical properties in terms of molecular weight, ring arrangement, aqueous solubility, adsorption-desorption properties, stability, volatility, oxidizability, and biodegradability. Unsubstituted PAHs are the most abundant. PAHs are produced by both natural processes and anthropogenic activities. They persist in nature and are widespread organic contaminants of air, water, soil, and sediment environments.

Most of the literature on PAH biodegradation describes aerobic laboratory batch studies. There are fewer reports on anaerobic biodegradation of PAHs. Dreyer, Konig, and Ringpfeil (1995) demonstrated biodegradation of 14 PAHs by a mixed bacterial culture under aerobic conditions. Microorganisms native to Buffalo River sediment, Buffalo, NY, were shown to degrade PAHs with fewer than five rings, and to partially degrade others at 30 °C under aerobic conditions in laboratory batch reactors (Jones, Araujo, and Rogers 1996). Numerous studies have shown that low-molecular-weight PAHs (up to three rings) are readily biodegraded as the sole source of carbon and energy and to some extent mineralized (Cerniglia 1984; Mueller, Chapman, and Pritchard 1989; Efrogmson and Alexander 1991; Christian, Pugh, and Clarke 1995; Pott and Henrysson 1995; Jee et al. 1998; Mohammed et al. 1998). Biodegradation of high-molecular-weight PAHs (four or more rings) by co-metabolism has also been reported (Sims and Overcash 1983; Sims et al. 1988; Keck et al. 1989). In co-metabolism, enzyme(s) produced for catabolism of the growth-supporting substrate also degrades a PAH that is not essential for growth.

Bacterial genera commonly reported to be involved in PAH biodegradation include *Acinetobacter*, *Aeromonas*, *Alcaligenes*, *Bacillus*, *Beijerinckia*, *Corynebacteria*, *Cyanobacter*, *Enterococcus Flavobacterium*, *Micrococcus*, *Mycobacterium*, *Nocardia*, *Pediococcus Pseudomonas*, *Rhodococcus*, and *Vibrio* (Wilson and Jones 1993; Mueller et al. 1994). White rot fungus, *Phanerochaete chrysosporium*, and another lignin-degrading fungus, *Phanerochaete sordida*, are also known to degrade a variety of PAHs (Hammel, Kalyanaraman, and Kent-Kirk 1986; Bumpus et al. 1989; Davis et al. 1993; May, Schroder, and Sandermann 1997).

Overall, the literature indicates that the higher molecular weight and more carcinogenic PAHs are less biodegradable than the lower molecular weight PAHs. Therefore, the least degradable fraction is generally subject to the most stringent cleanup standards. This presents some difficulty in achieving cleanup goals with bioremediation systems.

PCBs. PCBs encompass a class of chlorinated compounds that includes up to 209 variations or congeners with different physical and chemical characteristics. The PCB molecule consists of two benzene rings that are joined by a single covalent carbon:carbon bond with varying degrees of chlorination on each ring. PCBs were manufactured as PCB congener mixtures under the trade name Aroclors (National Research Council 1979). These very stable mixtures have widely varying fluidity, dielectric constant, vapor pressure, solubility, etc., and were thus suitable for many industrial applications. The original PCB mixtures, marketed as Aroclors, entered the environment

as waste transformer oils, hydraulic fluids, pesticide extenders, and plasticizers. Due to degradation and differential affinities of PCB congeners for various environmental compartments, PCBs in environmental samples are highly weathered by physical, chemical, and biological processes and rarely, if ever, match the original Aroclor composition (Duinker and Hillebrand 1983; Duinker, Hillebrand, and Boon 1983; Bush et al. 1985). Further, PCBs vary widely in toxicity (Bunyan and Page 1978; McFarland, Clarke, and Gibson 1986; Clarke, McFarland, and Pierce 1989).

Anaerobic biodegradation of PCBs in sediments via reductive dehalogenation, resulting in a lower number of chlorines, has been reported by several investigators (Bedard et al. 1986; Brown et al. 1987a; Brown et al. 1987b; Quensen, Tiedje, and Boyd 1988; Quensen, Boyd, and Tiedje 1990; Nies and Vogel 1990; Van Dort and Bedard 1991; Alder et al. 1993; Klasson et al. 1996; Natarajan et al. 1996; Natarajan et al. 1998; Wu, Bedard, and Weigel 1999). Anaerobic reductive dehalogenation is the only known biodegradation process for more highly chlorinated PCBs (Quensen, Boyd, and Tiedje 1990). Anaerobically, PCB dechlorinating bacteria dechlorinate higher chlorinated PCBs by taking off chlorines, leaving the biphenyl ring in place (Harkness et al. 1993). Aerobic biodegradation of PCBs has also been reported (Abramowicz, Brennan, and van Dort 1990; Harkness et al. 1993; Williams and May 1997; Tang and Myers 1998). Anaerobic conditions that leave PCBs more susceptible to biodegradation by aerobes can be produced in dredged material with relative ease.

Polychlorinated Dibenzo-*p*-dioxins (PCDDs) and Polychlorinated Dibenzofurans (PCDFs). Polychlorinated dibenzo-*p*-dioxins and their degenerated forms of polychlorinated dibenzofurans occur widely in the environment, especially in sediments (Clarke et al. 1994). These compounds were never intentionally manufactured, but are formed during chlorophenol and chlorinated herbicide synthesis, as toxic components of wood treating wastes derived from the use of pentachlorophenol, as by-products from bleach processes in the pulp and paper industries, and during incineration processes (Olie, Vermeulen, and Hutzinger 1977; Choudhary, Keith, and Rappe 1983; Rappe, Choudhary, and Keith 1986). Dioxins are a family of compounds composed of a triple ring structure consisting of two benzene rings that are interconnected by a pair of oxygen atoms. Furans are two benzene rings connected by one oxygen atom and one carbon:carbon bond. These substances, because of their hydrophobic nature, attach to soil and sediment particles. Storm runoff leads to contamination of sediments, particularly near chlorophenol production facilities.

Adriaens and co-workers (Adriaens and Grbic'-Galic' 1994; Adriaens, Fu, and Grbic'-Galic' 1995; Barkovskii and Adriaens 1995; Adriaens, Chang, and Barkovskii 1996; Albrecht, Barkovskii, and Adriaens 1999) and Beurskens et al. (1995) have shown that reductive dechlorination of PCDD and PCDF occurs in sediments, albeit very slowly. Presumably, once a sufficient number of chlorine atoms have been removed anaerobically, the remaining PCDD and PCDF would be biodegradable by aerobic microorganisms much like PCBs. In laboratory studies designed to simulate land treatment of dredged material, Myers and Tang (1998) reported no disappearance of PCDD and a statistically significant reduction of PCDF, although the loss of PCDF was attributed to volatilization.

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