

# **Bioaugmentation for Groundwater Remediation**

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**SERDP and ESTCP Remediation Technology Monograph Series**  
**Series Editor: C. Herb Ward, Rice University**



# Bioaugmentation for Groundwater Remediation

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# SERDP and ESTCP Remediation Technology Monograph Series

Series Editor: C. Herb Ward, Rice University

SERDP and ESTCP have joined to facilitate the development of a series of monographs on remediation technology written by leading experts in each subject area. This volume provides a review of the state-of-the-art on bioaugmentation for groundwater remediation. Previously published volumes include:

- *In Situ* Bioremediation of Perchlorate in Groundwater
- *In Situ* Remediation of Chlorinated Solvent Plumes
- *In Situ* Chemical Oxidation for Groundwater Remediation
- Delivery and Mixing in the Subsurface: Processes and Design Principles for *In Situ* Remediation

Additional volumes planned for publication in the near future include:

- Chlorinated Solvent Source Zone Remediation
- Processes, Assessment and Remediation of Contaminated Sediment



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## Preface

In the late 1970s and early 1980s, our nation began to grapple with the legacy of past disposal practices for toxic chemicals. With the passage in 1980 of the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA), commonly known as Superfund, it became the law of the land to remediate these sites. The U.S. Department of Defense (DoD), the nation's largest industrial organization, also recognized that it too had a legacy of contaminated sites. Historic operations at Army, Navy, Air Force, and Marine Corps facilities, ranges, manufacturing sites, shipyards, and depots had resulted in widespread contamination of soil, groundwater, and sediment. While Superfund began in 1980 to focus on remediation of heavily contaminated sites largely abandoned or neglected by the private sector, the DoD had already initiated its Installation Restoration Program in the mid-1970s. In 1984, the DoD began the Defense Environmental Restoration Program (DERP) for contaminated site assessment and remediation. Two years later, the U.S. Congress codified the DERP and directed the Secretary of Defense to carry out a concurrent program of research, development, and demonstration of innovative remediation technologies.

As chronicled in the 1994 National Research Council (NRC) report, "Ranking Hazardous-Waste Sites for Remedial Action," our early estimates on the cost and suitability of existing technologies for cleaning up contaminated sites were wildly optimistic. Original estimates, in 1980, projected an average Superfund cleanup cost of a mere \$3.6 million per site and assumed only around 400 sites would require remediation. The DoD's early estimates of the cost to clean up its contaminated sites were also optimistic. In 1985, the DoD estimated the cleanup of its contaminated sites would cost from \$5 billion to \$10 billion, assuming 400–800 potential sites. A decade later, after an investment of over \$12 billion on environmental restoration, the cost-to-complete estimates had grown to over \$20 billion, and the number of sites had increased to over 20,000. By 2007, after spending over \$20 billion in the previous decade, the estimated cost to complete the DoD's known liability for traditional cleanup (not including the Munitions Response Program for unexploded ordnance) was still over \$13 billion. Why did we underestimate the costs of cleaning up contaminated sites? All of these estimates were made with the tacit assumption that existing, off-the-shelf remedial technology was adequate to accomplish the task, that we had the scientific and engineering knowledge and tools to remediate these sites, and that we knew the full scope of chemicals of concern.

However, it was soon and painfully realized that the technology needed to address the more recalcitrant environmental contamination problems, such as fuels and chlorinated solvents in groundwater and dense nonaqueous phase liquids (DNAPLs) in the subsurface, was seriously lacking. In 1994, in the "Alternatives for Ground Water Cleanup" document, the NRC clearly showed that as a nation we had been conducting a failed 15-year experiment to clean up our nation's groundwater and that the default technology, pump-and-treat, was often ineffective at remediating contaminated aquifers. The answer for the DoD was clear. The DoD needed better technologies to clean up its contaminated sites, and better technologies could only arise through a better scientific and engineering understanding of the subsurface and the associated chemical, physical, and biological processes. Two DoD organizations were given responsibility for initiating new research, development, and demonstrations to obtain the technologies needed for cost-effective remediation of facilities across the DoD: the Strategic Environmental Research and Development Program (SERDP) and the Environmental Security Technology Certification Program (ESTCP).

SERDP was established by the Defense Authorization Act of 1991, as a partnership of the DoD, the U.S. Department of Energy, and the U.S. Environmental Protection Agency; its mission is “to address environmental matters of concern to the Department of Defense and the Department of Energy through support of basic and applied research and development of technologies that can enhance the capabilities of the departments to meet their environmental obligations.” SERDP was created with a vision of bringing the capabilities and assets of the nation to bear on the environmental challenges faced by the DoD. As such, SERDP is the DoD’s environmental research and development program. To address the highest-priority issues confronting the Army, Navy, Air Force, and Marine Corps, SERDP focuses on cross-service requirements and pursues high-risk and high-payoff solutions to the DoD’s most intractable environmental problems. SERDP’s charter permits investment across the broad spectrum of research and development, from basic research through applied research and exploratory development. SERDP invests with a philosophy that all research, whether basic or applied, when focused on the critical technical issues, can impact environmental operations in the near term.

A DoD partner organization, ESTCP, was established in 1995 as the DoD’s environmental technology demonstration and validation program. ESTCP’s goal is to identify, demonstrate, and transfer technologies that address the DoD’s highest priority environmental requirements. The program promotes innovative, cost-effective environmental technologies through demonstrations at DoD facilities and sites. These technologies provide a large return on investment through improved efficiency, reduced liability, and direct cost savings. The current cost and impact on DoD operations of environmental compliance is significant. Innovative technologies are reducing both the cost of environmental remediation and compliance and the impact of DoD operations on the environment, while enhancing military readiness. ESTCP’s strategy is to select laboratory-proven technologies with potential broad DoD application and use DoD facilities as test beds. By supporting rigorous testing and evaluation of innovative environmental technologies, ESTCP provides validated cost and performance information. Through these tests, new technologies gain end-user and regulatory acceptance.

In the 17–21 years since SERDP and ESTCP were formed, much progress has been made in the development of innovative and more cost-effective environmental remediation technology. Since then, recalcitrant environmental contamination problems for which little or no effective technology had been available are now tractable. However, we understand that newly developed technologies will not be broadly used in government or industry unless the consulting engineering community has the knowledge and experience needed to design, cost, market, and apply them.

To help accomplish the needed technology transfer, SERDP and ESTCP have facilitated the development of a series of monographs on remediation technology written by leading experts in each subject area. Each volume will be designed to provide the background in process design and engineering needed by professionals who have advanced training and five or more years of experience. The first volume in this series, *In Situ Bioremediation of Perchlorate in Groundwater*, met a critical need for state-of-the-technology guidance on perchlorate remediation. The second volume, *In Situ Remediation of Chlorinated Solvent Plumes*, addressed the diverse physical, chemical, and biological technologies currently in use to treat what has become one of the most recalcitrant contamination problems in the developed world. The third volume, *In Situ Chemical Oxidation for Groundwater Remediation*, provided comprehensive, up-to-date descriptions of the principles and practices of *in situ* chemical oxidation for groundwater remediation based on a decade of intensive research, development, and demonstration. The fourth volume, *Delivery and Mixing in the Subsurface: Processes and Design Principles for In Situ Remediation*, described the principles of chemical delivery and mixing systems, and their design and implementation for effective *in situ* remediation. Other volumes will follow on such topics as the remediation of DNAPL-chlorinated solvent source zones and remediation of



contaminated sediments. Additional volumes will be written as new remediation technologies are developed and proven to be effective.

This volume, *Bioaugmentation for Groundwater Remediation*, provides a review of the past 10–15 years of intensive research, development, and demonstrations that have been at the forefront of developing bioaugmentation into a viable remedial technology. In addition, both a primer on the basic microbial processes involved in bioaugmentation as well as a thorough summary of the methodology for implementing the technology is provided within this volume. It is our intention that this will serve as a valuable resource for environmental remediation professionals who seek to understand, evaluate, and implement bioaugmentation. Topics addressed in this volume include:

- A brief history and overview of bioaugmentation (Chap. 1).
- A detailed review of the discovery of *Dehalococcoides* and the development of reductive dechlorination of chlorinated solvents as a remedial technology (Chap. 2).
- The state-of-the-science for the production and handling of *Dehalococcoides* bioaugmentation cultures (Chap. 3).
- An overview of a decision process for determining whether to implement bioaugmentation with *Dehalococcoides* (Chap. 4).
- Design considerations for implementing bioaugmentation (Chap. 5).
- A summary of microbial monitoring options during bioaugmentation with *Dehalococcoides* (Chap. 6).
- A thorough review of the use of bioaugmentation for treatment of chemicals other than the more common chlorinated solvents (TCE and PCE), including DCE (Chap. 7), aerobic cometabolism of chlorinated solvents (Chap. 8), carbon tetrachloride (Chap. 9), and MTBE (Chap. 10).
- An analysis of cost considerations needed to evaluate whether bioaugmentation should be considered for the treatment of chlorinated aliphatic compounds in groundwater (Chap. 11).
- An assessment of important unknowns and uncertainties that impact the state-of-the-science that underpins bioaugmentation development and implementation. This chapter, written in consultation with a broad range of experts in the remediation field, should help guide the research agenda on bioaugmentation (Chap. 12).

Each chapter in this volume has been thoroughly reviewed for technical content by two or more experts in each subject area covered. The editors and chapter authors have produced a well-written and up-to-date treatise that we hope will prove to be a useful reference for those making decisions on remediation of contaminated sites, for remediation practitioners, and for those involved in development of advanced technology for the *in situ* remediation of contaminated groundwater.

SERDP and ESTCP are committed to the development of new and innovative technologies to reduce the cost of remediation of soil, groundwater, and sediment contamination as a result of past operational and industrial practices. We are also firmly committed to the widest dissemination of these technologies to ensure that our investments continue to yield savings for not only the DoD, but also the nation. In sponsoring this monograph series, we hope to provide the broader remediation community with the most current knowledge and tools available in order to bring these technologies to bear on the remediation of contaminated sites.

Jeffrey A. Marqusee, PhD, Executive Director, SERDP and ESTCP

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## About the Editors

### Hans F. Stroo

Dr. Stroo is a Principal Technical Advisor with HydroGeoLogic, Inc. He provides technical support on large remediation projects for private- and public-sector clients and has served as a technical advisor to the Strategic Environmental Research and Development Program (SERDP) and the Environmental Security Technology Certification Program (ESTCP) for over 10 years.

Dr. Stroo received his BS degrees in Biology and Soil Science from Oregon State University, his MS in Soil Science from West Virginia University, and his PhD in Soil Science (Soil Microbiology) from Cornell University.

He was formerly a Principal with Remediation Technologies, Inc. (RETEC). He has over 20 years of experience in the assessment and remediation of contaminated soil and groundwater, particularly in the development and use of *in situ* bioremediation.

Dr. Stroo has served on several expert review panels for SERDP, other government agencies, and private companies. Recently, he served as co-chair of the SERDP workshops on Remediation of Chlorinated Solvents in Groundwater and Remediation of Dense Nonaqueous Phase Liquid (DNAPL) Source Zones. He is coeditor of the SERDP-facilitated monographs, *In Situ Bioremediation of Perchlorate in Groundwater* and *In Situ Remediation of Chlorinated Solvent Plumes*.

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Dr. Leeson is the Environmental Restoration Program Manager for SERDP and ESTCP. She received her BS degree in Biology from Eastern Kentucky University and her PhD in Environmental Engineering from The Johns Hopkins University.

Dr. Leeson has been the Environmental Restoration Program Manager at SERDP and ESTCP since 2001. She was formerly a Research Leader at Battelle Memorial Institute and worked on the design and implementation of innovative physical/chemical and biological treatment technologies for site remediation and treatment of industrial/hazardous wastewater for 10 years prior to working at SERDP and ESTCP.

Dr. Leeson served as chairperson of the Fourth, Fifth, and Sixth International *In Situ* and On-Site Bioremediation Symposium and was the founding Managing Editor for *Bioremediation Journal* from 1996 to 1999 and the Editor-in-Chief from 1999 to 2001. She has developed and implemented several expert panel workshops on topics ranging from contaminant bioavailability to chlorinated solvent-contaminated groundwaters.

### C. Herb Ward

Dr. Ward holds the Foyt Family Chair of Engineering in the George R. Brown School of Engineering at Rice University. He is also Professor of Civil and Environmental Engineering and Ecology and Evolutionary Biology.

Dr. Ward has undergraduate (BS) and graduate (MS, PhD, MPH) degrees from New Mexico State University, Cornell University, and the University of Texas School of Public Health, respectively. He is a registered professional engineer in Texas and a Board Certified Environmental Engineer by the American Academy of Environmental Engineers.

He has been a faculty member at Rice University for 47 years, where he has served as Chair of the Department of Environmental Science and Engineering and the Department of Civil and Environmental Engineering and as the Founding Director of the University's Energy and Environmental Systems Institute. He has also served as Director of the U.S. Environmental

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Dr. Ward has been a member of the USEPA Science Advisory Board and served as Chair of the SERDP Scientific Advisory Board. He is the founding and current Editor-in-Chief of the international scientific journal *Environmental Toxicology and Chemistry*.

Dr. Ward received the Frederick George Pohland Medal for Outstanding Contributions to Bridging Environmental Research, Education, and Practice and the Brown and Caldwell Lifetime Achievement Award for Remediation in 2006, the Water Environment Federation Jack Edward McKee Medal for Achievement in Groundwater Restoration in 2007, and the Society for Industrial Microbiology and Biotechnology Charles Thom Award for bioremediation research in 2011.

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Dr. Aziz specializes in assessment and remediation of industrial and military sites impacted by a broad range of contaminants, including chlorinated solvents, polycyclic aromatic hydrocarbons (PAHs), benzene, toluene, ethylbenzene, and xylenes (BTEX) and perchlorate. She has led the development, design, and implementation of several bioremediation/bioaugmentation technologies for public and private sector clients and has developed environmental software (BIOCHLOR) distributed through the U.S. Environmental Protection Agency (USEPA).

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### Evan Cox

Mr. Cox is a Principal Remediation Scientist with Geosyntec Consultants in Guelph, Ontario, Canada. He received his BSc in Biology and his MSc in Microbiology from the University of Waterloo (UW) in Waterloo, Ontario, Canada. Mr. Cox has over 20 years of demonstrated experience in the development, feasibility evaluation, and application of innovative *in situ* remediation technologies, including monitored natural attenuation (MNA), enhanced *in situ* bioremediation (EISB), *in situ* chemical oxidation (ISCO), and metal-catalyzed reduction of chlorinated and recalcitrant chemicals in subsurface environments.

Mr. Cox has helped to pioneer the development of MNA and EISB technologies, including bioaugmentation, for remediation of chlorinated solvents, propulsion energetics (perchlorate, *n*-nitrosodimethylamine [NDMA]), and explosives (RDX, TNT) in porous media and fractured bedrock. He works with private sector interests and government research programs to develop innovative *in situ* treatment technologies and to demonstrate and validate their use at field-scale for widespread commercial use. As part of his *in situ* remediation research, development, and implementation work, he has authored over 30 professional publications and articles regarding

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### **Craig S. Criddle**

Dr. Criddle is a Professor of Civil and Environmental Engineering at Stanford University and Senior Fellow in the Woods Institute for the Environment. He received his BS in Civil and Environmental Engineering and his BA in Spanish from Utah State University in 1982, followed by an MS in Environmental Engineering in 1984. In 1990, he completed his PhD at Stanford University in Civil Engineering (Environmental Engineering and Science). His research focus is environmental biotechnology.

Dr. Criddle began his academic career in 1989 as a faculty member at Michigan State University (MSU). While at MSU, he served as Project Director for the Schoolcraft project, a field-scale test of bioaugmentation that involved faculty from diverse disciplines and institutions, staff scientists, students, consultants and outreach education to members of the public and officials in Michigan. The National Ground Water Association (NGWA) named it one of two Outstanding Remediation Projects for 2002. Since 1998, he has been a member of the Stanford faculty, serving as Associate Chair in 2003 and as Director of a bioremediation field project at the U.S. Department of Energy (DOE) Field Research Center in Oak Ridge, Tennessee (2000–2006). This project entailed a multi-year series of experiments focused on *in situ* sequestration of uranium in groundwater and involved faculty from several universities, DOE scientists, and many students. Dr. Criddle has over 100 peer-refereed publications and is coauthor with award-winning artist Larry Gonick of *The Cartoon Guide to Chemistry* (2006), a widely used supplementary text for high school and freshman chemistry.

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Dr. Edwards is internationally known for her work on anaerobic bioremediation, the application of molecular biology and metagenomics to uncover novel microbial processes, and the transition of laboratory research into commercial practice to develop bioremediation and bioaugmentation strategies for groundwater pollutants. Dr. Edwards and Geosyntec Consultants were recognized with the 2009 National Science and Engineering Research Council of Canada Synergy Award for their long-standing partnership that led to the development of KB-1<sup>®</sup>, a successful commercial microbial bioaugmentation culture. She has been inducted into the Canadian Academy of Engineering and is a Fellow of the American Association for the Advancement of Science.

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In 23 years at GE, his work has focused on the development of novel remedial solutions for polychlorinated biphenyls (PCBs), petroleum hydrocarbons, and chlorinated solvents. His current specialty is the design of passive bioremediation systems for chlorinated solvents in groundwater. He has served as the GE representative on the steering committee of the Remediation Technologies Development Forum (RTDF) Bioremediation Consortium, and more recently Project SABRE, and is a frequent contributor to journal articles and book chapters in the field of bioremediation.

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Prior to joining the faculty at ASU in 1994, he was a Senior Research Engineer at Shell Development in Houston, Texas. His research, teaching, and professional activities are focused on the development, evaluation, and optimization of *in situ* remediation technologies and on modeling and monitoring related to risk assessment. From 1993 to 2012, he was the editor of *Ground Water Monitoring and Remediation*.

### **Stephen S. Koenigsberg**

Dr. Koenigsberg is Vice President and Director of Advanced Remediation Technologies at Brown and Caldwell in Irvine, CA. He has worked as a team member on numerous projects involving *in situ* and on-site treatment protocols and has published over 150 technical articles focusing on bioremediation and environmental biotechnology. He received his BA from the City College of New York (CCNY) and his MS and PhD degrees from Cornell University.

Dr. Koenigsberg was a Founder of Regenesis, where he coinvented or managed the development of the company's major products including ORC<sup>®</sup>, HRC<sup>®</sup>, MRC<sup>®</sup> and Regenox<sup>®</sup>. Also during his tenure, Regenesis produced one of the first commercial *Dehalococcoides* products (BDI<sup>®</sup>) and associated deoxyribonucleic acid (DNA) probes (Census<sup>sm</sup>) through the work of Dr. Frank Löffler at the Georgia Institute of Technology. Upon retiring from Regenesis, Dr. Koenigsberg was a Partner at WSP and a Principal at ENVIRON. In 2010, he received a Lifetime Achievement Award from the Association for Environmental Health & Sciences (AEHS) Foundation. He is a member of several editorial and advisory boards and is an Adjunct Professor at California State University at Fullerton where he also serves on the Dean's Advisory Council.

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Mr. Krug has extensive experience in the development and evaluation of innovative technologies for remediation of soil, groundwater, and sediment at contaminated properties



for Fortune 500 companies, branches of the U.S. Department of Defense (DoD), and the National Aeronautics and Space Administration (NASA). The focus of his professional practice has been in taking new technologies for remediation of environmental contamination from the early development stage to successful field-scale application and adapting new and conventional technologies to solve client's real-world problems. He has been a pioneer in the development, demonstration, and application of zero-valent metal and biological processes for the treatment of chlorinated organic compounds. In 2007, he was inducted into the Space Foundation Technology Hall of Fame for his contributions to the development of emulsified nanoscale zero-valent iron technology for treatment of chlorinated solvent dense nonaqueous phase liquid (DNAPL) source zones.

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