A Discussion of the Effects of Thermal Remediation Treatments on Microbial Degradation Processes

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NOTICE

FOREWORD

This report discusses the potential effects—beneficial and detrimental—of thermal processes on contaminant degrading microorganisms in soil and groundwater. While it presents current research and evidence, there is a definite need to increase the discussion and exchange of knowledge between thermal and bioremediation experts. The goal of this paper is to create an awareness of the need for cooperative action in the environmental community for further research and development on the subject.

This report was prepared by a graduate student in environmental engineering from the Milwaukee School of Engineering during the summer of 2002. It has been reproduced to help provide federal and state project managers responsible for hazardous waste sites with information on the current status of this technology.

About the National Network for Environmental Management Studies (NNEMS)

NNEMS is a comprehensive fellowship program managed by the EPA's Office of Environmental Education. The purpose of the NNEMS Program is to provide students with practical research opportunities and experiences.

Each participating headquarters or regional office develops and sponsors projects for student research. The projects are narrow in scope to allow the student to complete the research by working full-time during the summer or part-time during the school year. Research fellowships are available in environmental policy, regulations, and law; environmental management and administration; environmental science; public relations and communications; and computer programming and development.

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The report is available on the Internet at www.clu-in.org.

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1 Introduction

Over the past two decades, there has been a search for the most economical, reliable, and timely technologies for soil and groundwater remediation of contaminated sites. Various technologies have been developed for combinations of contaminant type, soil type, and cleanup standard. One class of technologies that have been increasingly gaining popularity is in situ thermal treatment, which concentrates on source removal. In situ thermal treatment often can expedite the cleanup process for a site from years to months.

Although in situ thermal treatment increasingly has been chosen for remediation of contaminated sites, some are still skeptical of the effects of heating the subsurface. Some believe that by applying heat to the subsurface, indigenous microorganisms that may otherwise degrade the existing contaminants are eliminated. In effect, the concern is that excessive heat will sterilize the soil. This belief is based on the same principles that make sterilization of hospitals and pasteurization of dairy products possible. If this belief holds true, it could be argued that aggressive treatment options, such as thermal technologies, may actually be doing more harm than good. The argument is that the presence of contaminant degrading microorganisms justifies passive remediation because not only are aggressive thermal techniques more expensive than bioremediation or natural attenuation, they are detrimental to the soil as well.

On the other hand, thermal treatments are very timely in remediation capabilities, greatly accelerating cleanup. The application of a thermal technology can render a site safe and useful in less time than bioremediation or certainly natural attenuation. It is also argued that the application of heat may actually increase microbial degradation rates. It is well accepted that within a certain range of temperatures, a general rule of thumb is that degradation rates double for every 10 °C increase in temperature. This temperature range is dependant on temperature-toleration characteristics of the microbial consortia. Assuming thermal vendors operate systems within the optimal temperature range, microbial activity should increase with the increase in temperature, and contaminants will be degraded at an accelerated rate. The currently accepted cutoff temperature in which microbial activity ceases is typically 40 °C. [1] Still, little is known about the extent of microbial activity in soils at temperatures above 40 °C.

In situ thermal operating temperatures vary with the type of system. At least one vendor appears to operate under 40 °C when applying thermal remediation technologies, because they explicitly seek to take advantage of enhanced metabolic processes. Other thermal vendors, operating at or above 100 °C, intend to remove contaminants thermally. Because of this practice, it becomes important to investigate the effects on the microbial population at these elevated temperatures.

An important factor to consider is that thermal vendors typically target only source zones. The dissolved phase plume is not heated directly during thermal treatment, but areas of the plume may experience a temperature rise as heat is transferred through the subsurface. The extent of temperature increase in these areas is a function of the distance from the heated source zone. Further, there has been speculation of whether microbial activity exists in the source zone even prior to remedial treatment. A common belief is that contaminant concentrations in the source zone are too great for microorganisms to survive.

If the presence of high temperatures eliminates microbial growth in the subsurface, in situ thermal technologies could be detrimental. Thermal processes are generally limited to source zones, leaving natural attenuation processes in downgradient dissolved phase plume areas unaffected. Furthermore, in situ thermal technologies may not destroy subsurface microorganisms, but only temporarily alter metabolic processes, either positively or negatively. If this is true, the possible implementation of a treatment train using thermal technologies for source removal and bioremediation for dissolved phase reduction could become a promising remedial option to explore.

2 In Situ Thermal Remediation Technologies

In situ thermal technologies [2] increasingly have been chosen as an effective remediation technology for source removal. In situ thermal treatments combine vapor extraction with subsurface heating to increase the removal rate of contaminants. Heating the subsurface increases the volatility and solubility of contaminants; promotes rapid mass transfer, diffusion, and evaporation; lowers the viscosity of contaminants; and increases the rates of chemical reactions. [3] The combination of these factors produces a greater rate of contaminant removal.

The following sections describe the five main categories of in situ thermal treatment technologies: hot air injection, steam injection, electrical resistance, radio-frequency heating, and thermal conduction.

2.1 Hot Air Injection

Hot air injection increases the rate of removal by increasing the soil temperature through injection wells or injection through a large mixing auger. This process tends to dry the soil while heating, which can impair microbial degradation.

2.2 Steam Injection

Steam injection is an in situ thermal technology that not only heats the subsurface, but also creates a pressure differential to mobilize contaminants. Steam is injected into the subsurface to promote contaminant partitioning into the vapor and aqueous phases for removal. Steam injection is most appropriate in conditions of adequate permeability. Lower permeability zones can be treated if steam can be applied above and below the zone to allow heating by conduction.

2.3 Electrical Resistance

Electrical resistance (ER) heats the soil with an array of electrodes inserted into the contaminated area. An electrical current is applied to the electrodes and a voltage differential is created causing the soil to heat. As the soil is heated and dries, the electrical resistance increases. ER generally is limited to steam temperature.

2.4 Radio-Frequency Heating

Radio-frequency (RF) heating uses electrodes or antennae powered by a RF generator to heat the subsurface. The electrodes are placed either on the surface or in boreholes drilled into the contaminated area. RF heating can create subsurface temperatures well above those attainable by hot air or steam injection, typically as high as 150 to 200 °C. RF heating also significantly dries the soil during heating, possibly decreasing biodegradation rates.

2.5 Thermal Conduction

Thermal conduction heating uses heaters applied horizontally or vertically on or in the soil. Application of the technology involves the use of heater-only and heater-extraction wells. Most contamination is destroyed in situ. A small percentage is recovered and treated as a vapor at the surface. Heating elements can operate at temperatures as high as 750 to 800 °C. Remediation projects involving more volatile contaminants can operate at lower temperatures. The higher temperature applications are largely beyond the scope of this paper.

3 Soil Microbiology

Microorganisms in soil are one of the primary factors that make soil what it is. Microorganisms are the mechanism responsible for recycling the raw materials that make life possible. Microbial processes drive the cycles that replenish the earth's supply of oxygen, carbon dioxide, and water. [4] The microbial process of particular importance to this research is the metabolism of soil and groundwater contaminants, such as petroleum hydrocarbons, polycyclic aromatic hydrocarbons, and chlorinated hydrocarbons.

3.1 Microbial Metabolic Processes

Microorganisms metabolize organic compounds to obtain the energy stored in their arrangement of atoms. Microorganisms systematically degrade complex organic molecules that are rich in energy to simpler waste products that have less energy. Metabolic pathways that release stored energy by breaking down complex molecules are called catabolic pathways. In aerobic catabolic processes, the most prevalent and efficient catabolic pathways, oxygen is consumed as a reactant along with organic compounds. Oxygen acts as an electron acceptor in order to degrade complex organics to simpler compounds. The following is a generalized equation for an aerobic metabolic process. [5]

$$\text{Organic Compounds} + \text{Oxygen} \rightarrow \text{Carbon Dioxide} + \text{Water} + \text{Energy}$$

Anaerobic catabolic processes are a partial degradation of organics that occur without the help of oxygen. There are four different groups of bacteria that aid in the anaerobic degradation of organic compounds: hydrolytic bacteria, fermentative acidogenic bacteria, acetogenic bacteria, and methanogens. These four groups of microorganisms each play an important role in this anaerobic metabolic process. The following equation illustrates the metabolic pathway for the anaerobic metabolism of organics to methane. [6]

$$\text{Organic Material} \xrightarrow{\text{Hydrolytic Bacteria}} \text{Monomers} \xrightarrow{\text{Fermentative Acidogenic Bacteria}} \text{Organic Acids, Alcohols, Ketones} \xrightarrow{\text{Acetogenic Bacteria}} \text{Acetate} \xrightarrow{\text{Methanogens}} \text{Methane}$$

Chlorinated hydrocarbons are anaerobically degraded through biological reductive dechlorination. The following equation represents reductive dechlorination of trichloroethene (TCE) to ethene. [7]

$$\underset{\text{TCE}}{C_2Cl_3H} \rightarrow \underset{\text{cis/trans-1,2-DCE}}{C_2Cl_2H_2} \rightarrow \underset{\text{Vinyl Chloride}}{C_2ClH_3} \rightarrow \underset{\text{Ethene}}{C_2H_4}$$

When these processes are actively implemented to promote the biodegradation of harmful contaminants in soils, it is referred to as anaerobic bioremediation.

3.2 Microbial Temperature Characteristics

Microbial processes are very dependent on the characteristics of the environment in which the organism is found. Temperature is one characteristic that plays a large role in a microorganism's ability to function. The temperature of an environment can affect microbial processes in different ways. The temperature range will determine what types of microorganisms will be able to thrive in that particular environment, while the specific temperature within the range will affect the rate of microbial processes. Microorganisms are often classified on their optimal temperature range. The following table lists the classifications of microorganisms and their operating temperature range. [8]

Classification	Low	High
Psychrophilic	0 °C	20 °C
Mesophilic	20 °C	40 °C
Thermophilic	40 °C	80 °C
Hyperthermophilic	80 °C	>100 °C

Every microorganism has an optimal temperature at which the microorganism is at its most efficient. The further from this temperature the environment strays, the less productive the microorganism will be. The optimal temperature for a microorganism typically is around the middle of the operating temperature range (i.e., ~30 °C for mesophilic). An accepted rule of thumb is that biodegradation rates double for every 10 °C increase above the low temperature. Accordingly, biodegradation rates decrease above the optimum temperature. When temperatures increase above the operating range for a type of microorganism, a switch in the microbial consortia may be observed.

3.2.1 Microbial Activity at Elevated Temperatures

The assumption typically is made that microbial activity is at its height in the mesophilic temperature range compared to psychrophilic and thermophilic activity. In some cases, however, microbial activity is found to be comparably high at elevated temperatures. High microbial activity under thermophilic conditions can be attributed either to microbial acclimation by mesophiles to the new environment or to a switch in the microbial consortia prompted by the new environment. Two examples of high microbial metabolic activity under thermophilic conditions are petroleum refineries and composting piles. These examples are proof that microbial metabolic processes exist at elevated temperatures.

3.2.1.1 Petrophilic Thermophiles

Hyperthermophilic microorganisms are found in oil fields approximately 3,500 meters below ground surface. [9] Samples of produced fluids contain high concentrations of various anaerobic extremely thermophilic and hyperthermophilic microorganisms, indicating the presence of complex microbial communities in situ. Some of the bacteria found in these oil reservoirs are methanogens, and are able to use crude oil as a source of energy in anaerobic metabolism.

3.2.1.2 Composting Thermophiles

A compost consists of any readily degradable organic matter that is kept in a heap with sufficient nutrients and sufficient aeration to enable rapid microbial growth. In composting, there is an initial phase of rapid microbial growth on the most readily available organics. This phase is initiated by mesophilic microorganisms, which generate heat by their metabolism and raise the temperature of the compost inhibiting their own growth. The microbial consortia then switches to thermophilic microorganisms that metabolize organics and produce heat. The temperature of the compost is raised to 70 to 80 °C within a couple of days. This process eliminates mesophilic microorganisms and leads to a prolonged high-temperature phase that favors thermophilic microorganisms. Eventually, the compost cools and mesophilic microorganisms reappear to again take over metabolism of organics. [8]

3.2.2 Microbial Destruction

Although there is evidence that microorganisms can acclimate to temperature change, an increase in temperature is sometimes used to destroy harmful bacteria. An increase in temperature can be very effective in removing bacteria that are detrimental to health and safety. Two examples of the use of heat to effectively eliminate dangerous microorganisms are the processes of pasteurization and sterilization. These examples illustrate an immense sensitivity to heat that some microorganisms possess.

3.2.2.1 Pasteurization

Pasteurization is used to remove harmful bacteria from dairy products. [10] Specifically, it is the heating of food to a specific temperature for a specified period of time without allowing

recontamination of the product during the heat treatment process. The purpose is to make dairy products safe for consumption and to prolong the keeping quality of the products. The extent of microorganism inactivation depends on the combination of temperature and holding time. For example, the table below represents temperatures and holding times for Ontario pasteurization regulations. Temperature-time combinations other than ones listed must be approved by the regulatory body.

Product	Temperature	Holding Time
Milk	63 °C	30 min.
	72 °C	16 sec.
Frozen Dairy Dessert Mix	69 °C	30 min.
	80 °C	25 sec.
Milk-based Products	66 °C	30 min.
	75 °C	16 sec.

The combination of temperature and holding time is based on thermal-death time studies for the most heat-resistant pathogens. Therefore, pasteurization does not eliminate all microbial activity, only microbial activity harmful to health and safety.

3.2.2.2 Sterilization

Sterilization is the procedure of making some object free of live bacteria or other microorganisms, usually by heat or chemical means. [11] Sterilization is used in medical facilities to protect patients from infection. Heating an object is a very common method of sterilization. Objects are heated to temperatures between 100 and 140 °C in a very short time to destroy all microbial activity. Sterilization is very effective in eliminating harmful microorganisms, but is not permanent. Sterilized equipment must be protected from the environment to maintain sterility. Once a sterilized object is exposed to the environment, it becomes vulnerable to microbial activity once again.

4 Experience and Research

The concern for detrimental effects on microorganisms caused by elevated temperatures is a fairly new area of interest in the environmental community. For this reason, little as research appears to have been performed on the topic. One leader in research is the Pacific Northwest National Laboratory (PNNL), with fieldwork by PNNL's Current Environmental Solutions (CES). Other work has been in progress overseas by a Dutch electrokinetics vendor, Hak Milieutechniek B.V. The research currently available is only an introduction, and further investigations are necessary to examine the full effects of heat on various contaminant-degrading microorganisms. The research to date can be used as an example for necessary future research in the thermal market. Research performed to date is discussed in the following sections.

4.1 Observations by Current Environmental Solutions

Current Environmental Solutions (CES) is a vendor of Six Phase Heating™ (SPH™) and was formed by Battelle Memorial Institute to commercialize the SPH™ technology. CES recently began researching the possibilities of accelerated bioremediation by heating. In preparation, William Heath, the Chief Operating Officer of CES, revisited previous SPH™ projects to review the observed effects of heating on contaminant degradation. [12] Heath made some conclusions about the cause of in situ degradation based on data from previous projects. Among the possible causes of in situ degradation were (1) biodegradation by thermophilic consortia that are stimulated by heating, (2) thermally accelerated hydrolysis reactions, (3) oxidation-reduction reactions driven by a shift in thermo-chemical groundwater equilibrium, and (4) hydrous-pyrolysis oxidation under aerobic conditions. CES reviewed five SPH™ projects to examine the cause for in situ degradation of contaminants and biodegradation by thermophilic consortia to be a possible mechanism of in situ degradation at two of the five sites.

4.1.1 Cape Canaveral, Florida

CES performed a pilot-study of SPH™ as part of a multiple technology demonstration for the in situ remediation of TCE present as DNAPL at Cape Canaveral, Florida. Soil temperatures in the heated area ranged between an average of 80 to 120 °C. The duration of the heating lasted 11 months, from August 18, 1999, to July 12, 2000. The demonstration resulted in a 97 percent mass removal of DNAPL, at least 44 percent of which was removed through in situ degradation. Although CES could not identify specifically the mechanism responsible for degradation, soil sample analysis revealed a rise in microbial activity after heating.

4.1.2 SPH™ at a Commercial Fuel Storage Yard

In October 1998, CES applied SPH™ to a gasoline spill at a commercial fuel storage yard. Subsurface temperatures were held at boiling conditions for four weeks. Samples indicated a 98 percent reduction in benzene concentrations, and groundwater concentrations were reduced below the target level. A surprising reduction in total petroleum hydrocarbons, specifically diesel-range organics (TPH-DRO), prompted further soil analysis. Soil samples were analyzed for petrophilic microbiological activity before, 30 days after, and 90 days after heating. Results showed an increase in petrophilic microbiological activity 30 days after SPH™ treatment and a dramatic increase in activity 90 days after treatment. These results (Figure 1) suggest that the elevated temperatures

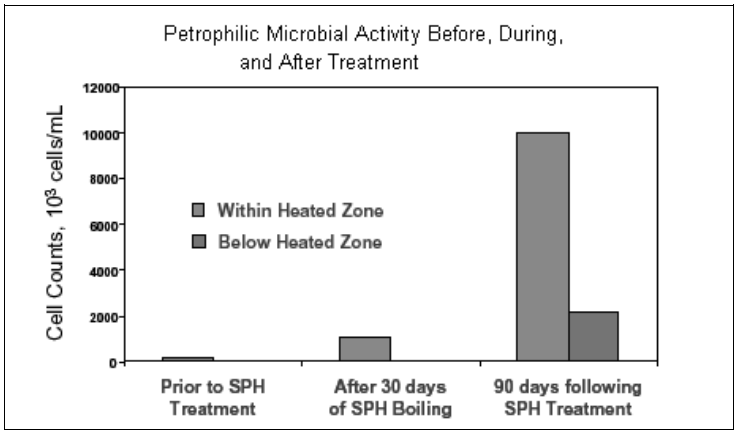

Figure 1. Microbiological activity before, during, and following six-phase heating treatment
Source: Current Environmental Solutions, 2001

did not destroy the microorganisms. It is likely that biodegradation at elevated temperatures contributed to contaminant removal.

4.2 Accelerated Bioremediation at Fort Wainwright, Alaska

The West Quartermaster's Fueling System (WQFS) in Operable Unit 5 (OU5) at Fort Wainwright, Alaska, underwent treatability studies for radio-frequency heating (RFH) and six-phase heating (SPH). [13] The most common in situ remediation technologies present at Fort Wainwright are soil vapor extraction (SVE) and air sparging (AS). Thermal technologies applied to SVE and AS can increase contaminant removal rates. The treatability studies for RFH and SPH were applied to assess thermal enhancements for effectiveness, operating parameters, and cost data. Both RFH and SPH were tested in order to assess the effectiveness of different soil heating technologies.

4.2.1 Site History

Fort Wainwright was placed on the National Priorities List (NPL) of the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) in August 1990. The remedial activities at the site are in place to bring the facility into compliance with state and federal regulations. The treatability studies are used to determine technologies that will bring the facility to compliance in the most cost effective manner.

The WQFS is divided into four areas, WQFS1 through WQFS4. The thermal treatability studies were conducted in area WQFS1 near three former fuel-pump islands. Soils in this area consisted of sand to sandy gravel. The site was contaminated with high concentrations of gasoline range organics (GRO) and diesel-range organics (DRO) from a few feet below the surface to approximately two feet below the water table. Benzene, toluene, ethylbenzene, and xylene (BTEX) and 1,2-DCA were the primary constituents present at the site.

4.2.2 Project Description

Three SVE/AS systems were constructed at WQFS1, two at each of the heated areas and one at an unheated control site. One heated site contained a RFH system and the other contained an SPH system. All three SVE/AS systems operated for 11 months. The RFH and SPH system operation were split into two periods: moderate-temperature heating and high-temperature heating. The moderate-temperature heating was to promote biodegradation of contaminants, and the high-temperature heating was to promote contaminant volatilization. Soil samples were taken and analyzed before heating, after moderate-temperature heating, and after high-temperature heating.

The study protocol included soil heating monitoring and SVE/AS remediation monitoring. Three objectives were set for soil heating monitoring: (1) change and uniformity in the vadose- and saturated-zone temperatures, (2) electrical energy use and heating efficiency, and (3) assessment of design parameters for full-scale implementation and comparison of SVE/AS with and without heating enhancements. Six objectives were set for SVE/AS remediation monitoring: (1) potential

increase in biodegradation rates associated with the increased soil temperature from near 0 to about 30 °C; (2) changes in soil moisture content attributable to SVE alone and to SVE with soil heating, and assessing potential impacts on biodegradation; (3) SVE and AS radius of influence; (4) assessing the increase in volatilization during moderate and high temperature operation; (5) estimating SVE remediation times for vadose zone at ambient, moderate, and high temperatures; and (6) measuring hydrocarbon concentrations in site soils to assess remediation levels and support theoretical calculations.

4.2.3 PROJECT OPERATION

Moderate-temperature operation was kept between 15 and 40 °C, temperatures found by CH_2M HILL to be beneficial for biodegradation rates. High-temperature operation included temperatures ranging from 60 to 100 °C. The moderate-temperature RFH system operated for 351 days (March 26, 1998, until March 11, 1999). High-temperature RFH operation began March 12, 1999 and ran until May 13, 1999 (62 days). The moderate-temperature SPH system operated for 98 days (March 26 to July 1, 1998). The high-temperature SPH system was in operation 55 days (July 2 to August 25, 1998).

4.2.4 BIODEGRADATION MONITORING RESULTS

Before the field study began, laboratory column studies were conducted on soil from WQFS1 to predict the effects of heat on biodegradation rates. Laboratory studies showed that although biodegradation rates were low at temperatures less than 5 °C, rates more than doubled when soil was heated to 10 to 15 °C. The studies also indicated that the optimum temperature for microbial consortia existing in the soil at WQFS1 is about 20 °C, and biodegradation rates declined at temperatures above 30 °C. Figure 2, below, graphically shows biodegradation results for the (a) RFH site and the (b) SPH site.

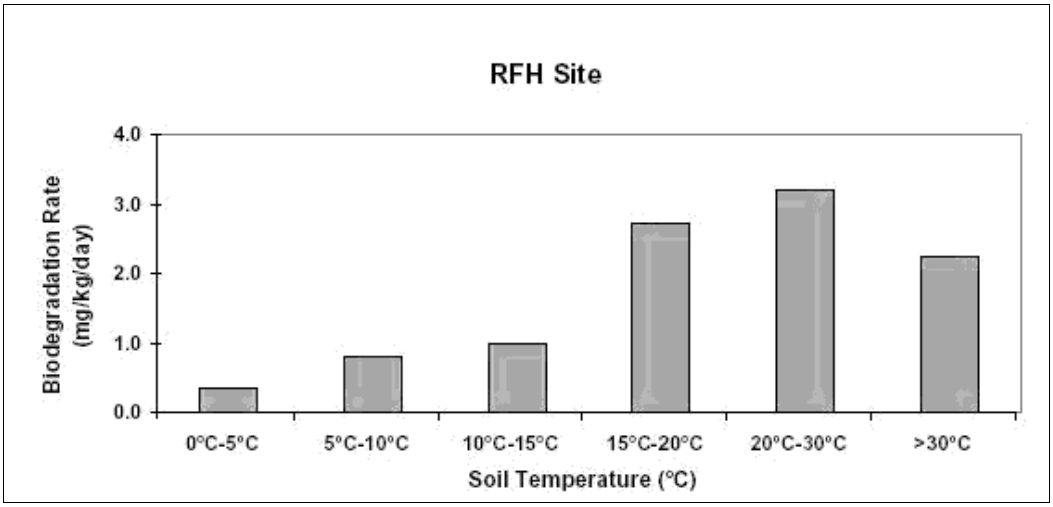

Figure 2(a). Biodegradation results from RFH site
(Source: CH2M Hill, 1999)

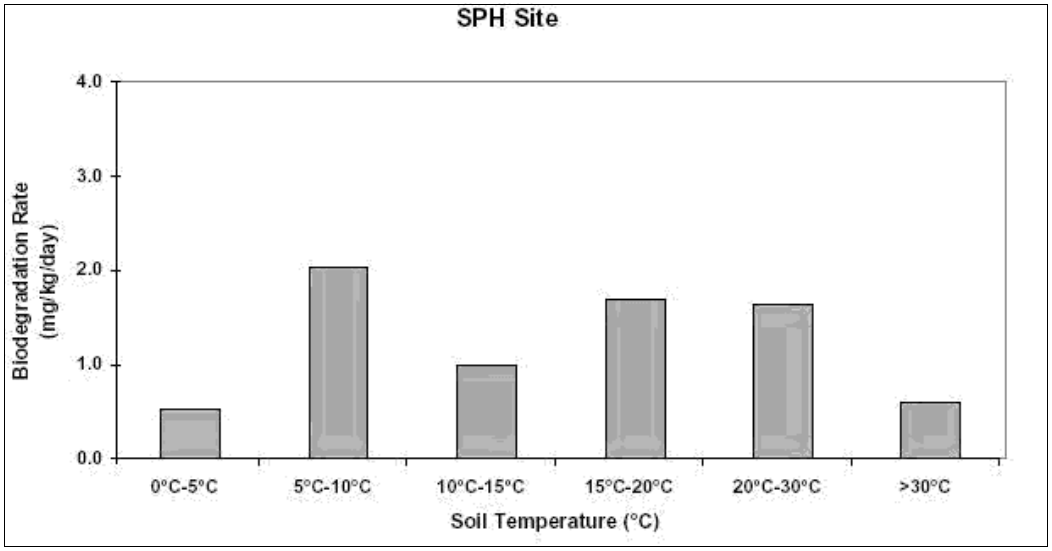

Figure 2(b). Biodegradation results from SPH site
(Source: CH2M Hill, 1999)

Monitoring of biodegradation rates was accomplished by respiration tests at the unheated control site, the RFH site, and the SPH site. Results showed a varying increase in biodegradation rates for the RFH and SPH sites heated to 10 to 25 °C. CH_2M Hill developed a model to quantify the effects of oxygen diffusion on resulting biodegradation rates. Using the model to correct for oxygen diffusion, results were consistent that biodegradation rates increased with an increase in soil temperature above 5 °C up to about 30 °C.

4.3 PNNL Study On Biodegradation of PAHs and Diesel

PNNL's Marine Sciences Laboratory in Sequim, Washington, conducted a research project on thermophilic biodegradation for PAHs and diesel in soil.[14] The purpose of the study was to assess whether biodegradation of hydrocarbons could occur following thermal remediation treatments. During thermal treatments soil vapors can travel through the cooler vadose zone and petroleum may condense, contaminating the vadose zone. PNNL investigated whether themophilic bioremediation of hydrocarbons will rectify this process and remove contaminants relocated to the vadose zone.

4.3.1 PROJECT DESCRIPTION

PNNL simulated a contaminated area on a laboratory scale by filling buckets with spiked sand, saturating the sand, then applying a layer of dry sand on top. The saturated zone in each bucket was spiked with either diesel fuel or a combination of six PAHs dissolved in toluene. The lower saturated zone was then heated to 110 °C in an oil bath for 90 minutes to simulate in situ thermal treatment. Water was siphoned to the heated sand to maintain levels lost by evaporation. This created high contaminant concentrations in the vadose zone because, as water was restored, the diesel NAPL was displaced from saturated soil and smeared throughout the overlaying vadose layer.

Following thermal treatment, soil samples were analyzed at 25, 50, and 70 °C for microbial activity. The samples were fertilized and aerated prior to analysis. A control column maintained at 70 °C was unfertilized and poisoned to eliminate microbial populations. This column was used to estimate hydrocarbon losses due to volatilization. Soil columns were analyzed at 0, 1, and 3 months for diesel hydrocarbon and PAH concentrations.

4.3.2 RESULTS

Light and medium diesel hydrocarbons and low-molecular weight PAHs were completely volatilized in the abiotic control at 70 °C. Theoretical estimates of volatilization losses indicated that these contaminants would completely volatilize at 50 and 70 °C. Therefore, it was assumed that any reduction in concentrations of light and medium diesel hydrocarbons and low-molecular weight PAHs at 50 and 70 °C is the result of volatilization and not biodegradation.

The rates of reduction for the heavier diesel hydrocarbons and the less-volatile PAHs were much faster in the biotic 70 °C sample than in the poisoned control 70 °C sample, suggesting that thermophilic biodegradation of contaminants had occurred. Biodegradation of heavy diesel hydrocarbons was shown to be relatively slow in the 25 and 50 °C samples compared to the 70 °C sample. Further, the slow rate of removal in the poisoned control indicated that the rapid reduction in the 70 °C sample is largely due to biodegradation. There was an increase in the rate of removal in the poisoned 70 °C sample after one month of treatment that may be indicative of a decrease in the effectiveness of the microbial poison and an increase in thermophilic biodegradation (Figure 3).

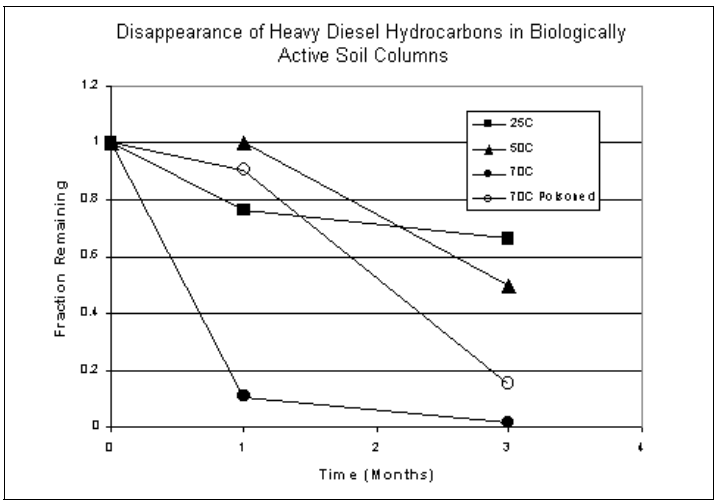

Figure 3. Thermophilic biodegradation of PAHs (Source: PNNL, 2002)

Soil samples were also analyzed for total aerobic heterotroph counts. For PAH contaminated soils, the highest counts were found in the 50 °C sample, while counts in diesel contaminated soils were greatest in the 25 °C samples. Relatively high counts were found in the poisoned 70 °C samples indicating a decrease in the effectiveness of the poison after the three-month period.

4.4 Future Research at Fort Lewis East Gate Disposal Yard

PNNL has been working to develop cleanup strategies, technology performance criteria, and remediation performance and compliance monitoring specifications associated with in situ thermal remediation technologies at the East Gate Disposal Yard (EGDY) in Fort Lewis, Washington. [15] The emphasis of the project is bioremediation and combinations of thermal treatment and bioremediation technologies. PNNL is working to deploy a cleanup strategy in

which in situ thermal treatment is used to target the large quantities of DNAPL at the site followed by bioremediation used to remove dissolved contaminants.

4.4.1 Project Objectives

The main objective of the project is to explore bioremediation technology approaches that would save time and reduce costs. PNNL intends to assess what actions are necessary in order to get the best advantage from and what factors affect the implementation of bioremediation during and after thermal treatment. Specifically, PNNL will determine whether dechlorination of TCE and daughter products will occur and what amendments are needed for dechlorination at temperatures above 50 °C. PNNL also intends to identify which factors are important in applying a combination of bioremediation and thermal treatment.

4.4.2 Project Description

The project will be split into two tasks. The initial evaluation of bioremediation issues is scheduled to begin in September 2002 and be completed in January 2003. The project is scheduled to be completed in December 2004.

4.4.2.1 Dechlorination Activity as a Function of Temperature

The first task is focused on the dechlorination activity as it relates to temperature. The purpose of this task is to address whether dechlorination will occur at temperatures above 50 °C. Task 1 also will investigate what amendments are needed for efficient microbial dechlorination at elevated temperatures. To accomplish this task, PNNL will revisit previous remediation work at EGDY. Review of "Reductive Anaerobic Biological In Situ Treatment Technology" (RABITT) studies performed at EGDY will provide insight to the implementation of bioremediation approaches. Laboratory tests will then be performed to determine the rates of dechlorination at elevated temperatures.

4.4.2.2 Bioremediation Implementation Issues

The second task addresses the issues related to implementation of bioremediation technologies at the EGDY source area. PNNL plans to develop a conceptual model to serve as a technical basis for a source area treatment strategy. The model will provide a basis for determining site-specific issues that are important to understand for application of candidate remediation approaches. PNNL proposes to use the conceptual model to evaluate the technical issues for bioremediation approaches. Evaluation will include assessment of laboratory results, field data, and other information relevant to bioremediation that may suggest that specific technical approaches would provide the most benefit for remediation of the EGDY source area.

4.5 Electro-BioReclamation

An electrokinetics vendor, Geokinetics International Inc., has recorded observations about microbiological activity following the application of electrokinetic remediation technology. In the Netherlands, Geokinetics was acquired by Hak Milieutechniek b.v, who manages the market in Europe for Geokinetics. The company employs an electrokinetic remediation technology called "electro-reclamation." The main principle of this technology is to move contaminants through the subsurface with electric current. Heat is a result of the process and subsurface temperatures rise accordingly.

4.5.1 ELECTRO-RECLAMATION EXPERIENCE

During numerous electro-reclamation projects, Geokinetics observed accelerated biodegradation rates as temperatures rose. [16] They reported that gradual heating of the soil up to 80 °C resulted in abundant biological activity at significantly higher levels than in the mesophilic range. After the heating has ended, Geokinetics observed the microbial populations adaptation to cooling temperatures and implemented a further increase in biological activity through periodic nutrient injections of nitrogen and phosphate. Geokinetics refers to this process as electro-bioreclamation, a derivative of electro-reclamation that combines thermal treatment for source removal and bioremediation as a polishing technique.

4.5.2 HAK MILIEUTECHNIEK B.V. RESEARCH

In 1999, two graduate students at the Van Hall Instituut in Leeuwarden completed a research project on the impact of high temperatures on the anaerobic biodegradation of chloroethenes on assignment of Hak Milieutechniek B.V. Schweitzer and Tuil [17] conducted pilot studies on a site contaminated with PCE, TCE, and daughter products.

4.5.2.1 Project Description

Remediation of the site began in 1990 with pump-and-treat. The method became unproductive, as the rate of removal of free product steadily decreased. For this reason, electro-reclamation was employed at the site in the summer of 1999. Schweitzer and Tuil set two objectives for their research: (1) to evaluate the investigation and remediation activities, which have been done in the past; and (2) to investigate the effect of temperature on the anaerobic biological degradation of chloroethenes. Only the second objective relates to the scope of this paper.

Schweitzer and Tuil studied microbial activity at temperatures of 10, 35, 62, and 70 °C. Three samples were heated slowly for each temperature previously mentioned, and one abiotic sample served as a control. A known concentration of PCE was added to each of the samples after heating. The samples were then monitored for 45 days for concentration of PCE and daughter products.

4.5.2.2 Results and Conclusions

Schweitzer and Tuil discovered that bacteria were present in all samples. Groundwater temperatures at this site are typically around 13 °C; however, bacteria were present at temperatures of 62 and 70 °C. Schweitzer and Tuil found that the dominant consortia varied with temperature. As expected, the greatest microbial activity was found in the 35 °C sample. This activity is consistent with mesophilic microbial activity. Microbial counts in the 62 and 70 °C samples were relatively low compared to those in the 10 and 35 °C samples.

Results showed that significant degradation had not occurred in any of the samples. This could be due to the short duration of the experiment. The samples were only analyzed up to 45 days, and degradation rates would likely increase over a longer duration had the experiment continued. Since degradation was nonexistent in the samples, the microbial activity present in the samples was assumed by Schweitzer and Tuil to exclude chloroethene degrading bacteria.

5 Discussion

Based on the information sources previously identified, the general state of knowledge on this topic appears to be limited. Although some research exists on the effects of thermal treatments on microorganisms, little can be concluded about how exactly microorganisms in the subsurface are affected by elevated temperatures. The large extent of uncertainty on this subject is an indication that more research is needed in order to fully understand the state of subsurface conditions following in situ thermal treatment.

5.1 Summary of Findings and Research

From the evidence presented above, a few general conclusions can be made. The following conclusions are discussed in detail below:

- Thermophilic microorganisms can degrade petroleum hydrocarbons;
- Change in subsurface temperature promotes change in microbial activity over time; and
- Possibility exists for a treatment train of thermal treatment for source removal and bioremediation for the dissolved phase.

5.1.1 Thermophilic Microorganisms Can Degrade Petroleum Hydrocarbons

Results from the PNNL study on thermophilic biodegradation of PAHs and diesel showed that thermophilic microorganisms are capable of degrading petroleum hydrocarbons. The rate of degradation for heavy diesel hydrocarbons and high molecular weight PAHs in biotic 70 °C samples was found to be significantly greater than rates in abiotic poisoned 70 °C samples. This suggests that thermophilic microorganisms had a significant impact on the degradation of these particular contaminants. Lighter diesel hydrocarbons and low molecular weight PAHs could not be analyzed for thermophilic biodegradation because these compounds were completely volatilized at higher temperatures.

5.1.2 Change in Subsurface Temperature Promotes Change in Microbial Activity Over Time

This conclusion can be seen in most of the cases presented above. The results from the Fort Wainwright research illustrate this conclusion rather well. At both the RFH and the SPH sites at Fort Wainwright, the biodegradation rates were observed to change as a function of temperature. The disagreement in the optimum temperature for contaminant degrading microbial activity could be a result of experimental error. In both cases, however, the biodegradation rates in the 20 to 30 °C range were relatively high in comparison to rates at other temperatures, including ambient temperatures. This is consistent with the optimal temperature range for mesophilic activity. At the SPH site, the two peak biodegradation rates were consistent with optimal temperatures for both psychrophilic (~10 °C) and mesophilic (~30 °C) activity, with a decrease in rates at other temperatures. This conclusion can also be seen, although to a lesser degree, in the CES findings at the Fuel Storage Yard. From Figure 1 it can be seen that the petrophilic cell counts dramatically increased following thermal treatment.

5.1.3 Possibility of Treatment Train of Thermal Treatment and Bioremediation

All of the cases discussed above show the possibility for a treatment train using thermal treatment for source removal and bioremediation for dissolved phase reduction. The use of this treatment train could be a technically and economically feasible way to reduce contaminant concentrations to Maximum Contaminant Levels (MCLs). For thermal technologies, remediation costs increase as the treated volume increases. As a result, in situ thermal treatment typically is applied to the source zone and not the dissolved phase area, or plume.

Bioremediation can be a very economical way to remove dissolved contaminants from the subsurface. Accordingly, bioremediation can be implemented as a polishing technique. From the cases discussed above, it is apparent that bioremediation has potential to follow thermal treatment, and that thermal treatment may even enhance the biodegradation rates of contaminants. The combination of thermal treatment for source removal and bioremediation for dissolved phase reduction could significantly reduce remediation costs and energy consumption at a contaminated site.

5.2 Possible Effects of In Situ Thermal Treatment on Microbial Degradation

There are many hypotheses about what effects in situ thermal treatment have on microbial degradation. One hypothesis is that elevated subsurface temperatures will inhibit, if not completely halt, microbial growth. Another is that heating the subsurface will promote a switch in the microbial consortia not only during the heating phase, but also during the cooling process. With the research currently available, neither of these scenarios can be confirmed or denied.

On the topic of whether elevated temperatures inhibit microbial growth, perhaps how elevated temperatures affect the growth of certain species should be researched on a site specific basis. Some species of microorganisms are much more sensitive to environmental conditions than others, therefore certain extreme climates can only harbor few strains of bacteria. Whether the

indigenous microorganisms at a site are particularly temperature sensitive will impact the possibility of implementation of bioremediation following thermal treatment.

Petrophilic microorganisms, for example, are known to thrive in high temperatures, as previously discussed. [18] This can be at least partially be accredited to the enhanced bioavailabilty of petroleum products at elevated temperatures resulting from increase in the solubility and decrease in the viscosity of the petroleum. To illustrate, the United Arab Emirates University researched the potential for the use of thermophilic bacteria in bioremediation of petroleum contaminants. While results were fundamentally inconclusive, it appeared that microbial cell counts were more dependent on the amount of crude oil present than on the temperature of the medium. When crude oil was readily available for metabolism, microbial growth rose. [19]

Little is known about the thermal effects on dechlorinating microorganisms. Dechlorinating microorganisms are typically known to be mesophilic and non-spore forming, which means they do not possess the capability to survive high temperatures. [17, 20] No research was found, however, on the possible existence of thermophilic dechlorinating bacteria. Clearly, more research on the effects of elevated temperatures on dechlorinating bacteria is needed.

Even with the possibility of inhibited growth of contaminant degrading species at high temperatures, bioremediation following in situ thermal treatment could still be a practical application for remediation. In the PNNL research on thermophilic bioremediation of PAHs and diesel, samples were fertilized and aerated following thermal treatment. The fertilization and aeration of samples could have provided the environment needed for surviving microorganisms to overcome any negative effects caused by elevated temperatures.

The flow of groundwater into the heated zone may offset any microbial activity inhibited by increased temperatures. Groundwater flowing into the treated zone would carry additional microorganisms where populations may have been decreased during thermal treatment. The movement of microorganisms into the treated area would eventually rejuvenate any areas that lacked significant microbial growth caused by elevated temperatures. Heated groundwater flowing from or through the thermally treated area could also promote a rise in biodegradation rates in remaining plume areas downgradient from the source area by increasing temperatures. Some areas of the plume would experience temperatures increased to the optimal range for mesophilic and thermophilic microbial activity, essentially creating a zone of enhanced biodegradation.

6 Recommendations

Based on the information currently available, it is apparent that a need for further research exists. Suggested topics for research include:

- Long-term analysis of microbial activity following thermal treatment;
- Biological characterization studies on contaminant degrading microorganisms;
- Determination of microbial consortia present in soils at various stages of thermal treatment;
- Pilot tests on the implementation of combination thermal treatment and bioremediation; and

- Thermal treatments effects on site conditions necessary for microbial degradation (i.e. moisture content, oxygen content, nutrient content).

It is necessary to explore these research topics in order to gain a true understanding of the effects of thermal treatments on microbial degradation processes. The combination of thermal treatment for source removal and bioremediation as a polishing technique has potential to be used as an effective treatment train in the near future. Before this combination can be implemented, however, a better knowledge of the degradation processes affected by thermal activity must be acquired.

7 Abbreviations

AS	Air Sparging
BTEX	Benzene, Toluene, Ethylene, and Xylene
CERCLA	Comprehensive Environmental Response, Compensation, and Liability Act
DCA	Dichloroethane
DCE	Dichloroethylene
DNAPL	Dense Non-Aqueous Phase Liquid
DRO	Diesel Range Organics
EDB	Ethylene Dibromide
GRO	Gasoline Range Organics
MCL	Maximum Contaminant Level
NAPL	Non-Aqueous Phase Liquid
NPL	National Priorities List (Superfund)
PAH	Polycyclic Aromatic Hydrocarbon
PCE	Perchloroethylene
RABITT	Reductive Anaerobic Biological In Situ Treatment Technology
RFH	Radio Frequency Heating™
SPH	Six Phase Heating™
SVE	Soil Vapor Extraction
TCA	Trichloroethane
TCE	Trichloroethylene
TPH	Total Petroleum Hydrocarbons

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