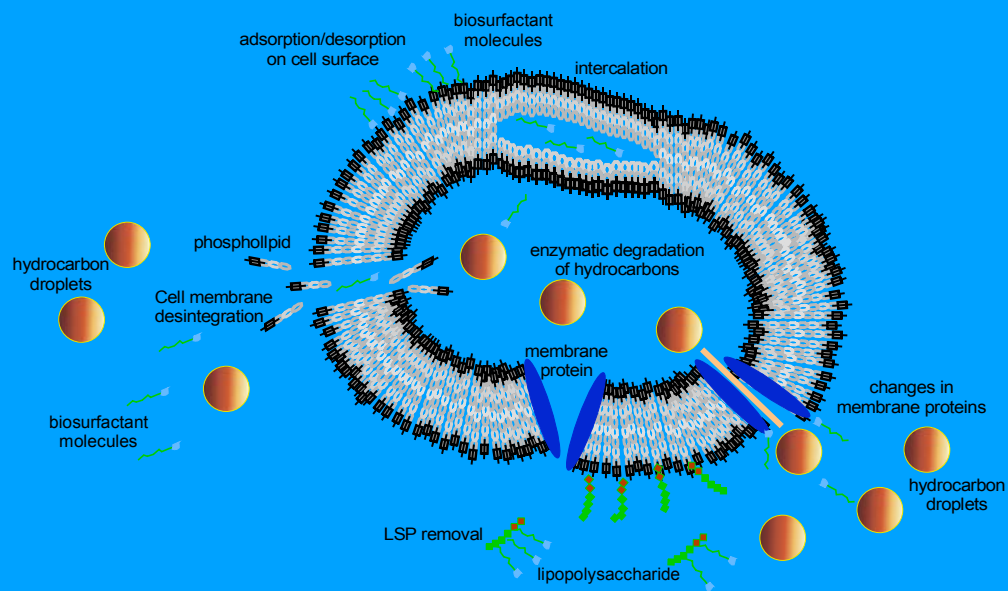


BIOREMEDIATION OPTIMIZATION PRIMER



Mark D. Ryckman
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Technical Publications

ABSTRACT

BIOREMEDIATION OPTIMIZATION PRIMER

Mark D. Ryckman, P.E., BCEE
Larry K. Seabolt, Jr., CHMM
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Bioremediation is an efficient technology when compared to alternative technologies. The two major advantages are lower capital cost and the ability to perform the task insitu. Beginning around 2000, enhanced biodegradation, or biostimulation, became widely accepted as a leading treatment technology. Many effective biostimulation supplements have been demonstrated and include the addition of surfactants, nutrients, enzymes, electron donors, electron receptors, and carbon substrates (1).

The purpose of this treatise is to 1) provide an understanding of bioremediation's key mechanisms, 2) provide optimization techniques and sample delivery methods, 3) present work flow diagrams for different matrix applications, and 4) present case histories for petroleum hydrocarbon bioremediations in soil and groundwater (Figure 17), railroad ballast (Figure 18), and gravel/soil at power plants and electrical substations (Figure 19) for a select non-toxic bioremediation accelerant, HC-2000.

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Remtech Engineers, May 2019

1. INTRODUCTION

Completing a significant number of soil and groundwater remediation projects to very low limits has been technically and financially unachievable. Over 90% of the superfund sites remain uncompleted. Underlying issues include: lack of understanding of the complex engineering and scientific subsurface biological, chemical, and physical processes; difficulty in quantifying contaminant mass transport through a variable and limiting subsurface cross section; underestimating the impact of contaminant sorption and rebound (back-diffusion); lack of experience with recalcitrant environmental contaminants such as Dense Non-Aqueous Phase Liquids (DNAPLs), fuels, and chlorinated solvents; and lack of funding (1, 2, 3).

Risk-based solutions that leave some of the contamination behind, while protecting public health and safety, are becoming more prominent and cost-effective. Solutions often involve removal, control, containment, or concentration reduction of the mobile/soluble phase to facilitate natural biological attenuation. This paper deals primarily with bioremediation of refined petroleum hydrocarbons rather than crude oil in fresh water environments. However, some of the same approaches may be utilized on crude oil in saline settings.

Bioremediation is an efficient technology when compared to alternative technologies. The two major advantages are lower capital cost and the ability to perform the task insitu. Beginning around 2000, enhanced biodegradation, or biostimulation, became widely accepted as a leading treatment technology. Many effective biostimulation supplements have been demonstrated and include the addition of surfactants, nutrients, enzymes, electron donors, electron receptors, and carbon substrates (1).

The purpose of this treatise is to 1) provide an understanding of bioremediation's key mechanisms, 2) provide optimization techniques and sample delivery methods, 3) present work flow diagrams for different matrix applications, and 4) present case histories for petroleum hydrocarbon bioremediations in soil and groundwater (Figure 17), railroad ballast (Figure 18), and gravel/soil at power plants and electrical substations (Figure 19) for a select non-toxic bioremediation accelerant, HC-2000.

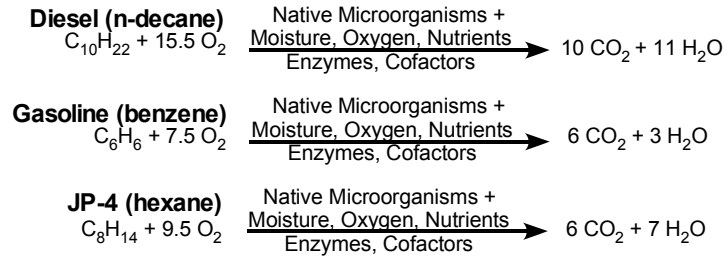
2. UNDERSTANDING BIOREMEDIATION

Biodegradation is defined as destruction of a chemical by the action of living organisms (4). Bioremediation uses microbial metabolism in the presence of optimum environmental conditions and sufficient nutrients to breakdown contaminants, notably petroleum hydrocarbons. Bioremediation efficacy hinges on site specific requirements such as the availability of sufficient quantities of microorganisms capable of degradation, nutrient availability to support microbial growth and proliferation, contaminant concentration and type, contaminant location, and environmental parameters including temperature in combination with duration of exposure.

Bioremediation involves the production of energy in a redox reaction within microbial cells. These reactions include respiration and other biological functions needed for cell maintenance and reproduction. A delivery system that provides one or more of the following is generally required: an energy source (electron donor), an electron acceptor, and nutrients. Different types of microbial electron acceptor classes can be involved in bioremediation, such as, oxygen, nitrate, manganese, iron (III), sulfate, or carbon dioxide, and the corresponding redox potentials. Redox potentials provide an indication of the relative dominance of the electron acceptor classes (5).

Cometabolism is another mechanism involved in the breakdown of contaminants. Cometabolism is defined as a biotransformation when microorganisms use other carbon sources as their primary substrate rather than the contaminant to produce enzymes that degrade the contaminant.

Simplified Microbial Aerobic Weathering/Mineralization of Petroleum Hydrocarbons



2.1 Role of Native Microorganisms

The majority of soil and groundwater petroleum hydrocarbon contaminated sites contain a sufficient consortium (mixture) of microorganisms (bacteria, fungi, yeast) capable of degrading most contaminants (5, 6, 7, 8). Multiple native organisms are present in the aerobic, facultative, anaerobic, methanogenic, and fermentative zones that all contribute to the mineralization of organic contamination. No single organism can complete the process. What is generally missing are the complete conditions necessary for indigenous species to function effectively. Specific requirements include nutrients, growth substrate inducers, electron donors, and electron acceptors. Most of the native bacterial species in soil remain unidentified due to limitations in existing microbial identification methodologies. The mere presence of documented petroleum degraders may not be enough to ensure degradation. Less than 1% of environmental isolates are cultural and therefore do not accurately represent the actual community of hydrocarbon degraders (8).

Native soil populations capable of degrading petroleum hydrocarbons have been reported to range from 10 to 20% of all soil heterotrophic bacteria. In railroad ballast, 59 to 94% of all organisms are petroleum hydrocarbon degraders. Soil particles containing heterotrophs are found in lower concentrations in groundwater and surface water (2×10^4 cells/ml to 2×10^6 cells/ml) than in soil (2×10^7 cells/gm) (9), but provide enough metabolic and synthetic activity to digest petroleum hydrocarbons. Approximately 67% of all bacteria in groundwater sediment are attached to the silt and clay fraction (3).

2.2 Microbial Assimilation

Native communities, which are exposed to hydrocarbons or pollutants, become adapted, exhibiting selective enrichment and genetic changes where contamination becomes their source of food. The adapted microbial communities can respond to the presence of hydrocarbon pollutants within hours and exhibit higher biodegradation rates than communities with no history of hydrocarbon contamination (Figure 1)(5).



Figure 1: Microbial Adaption to Contamination

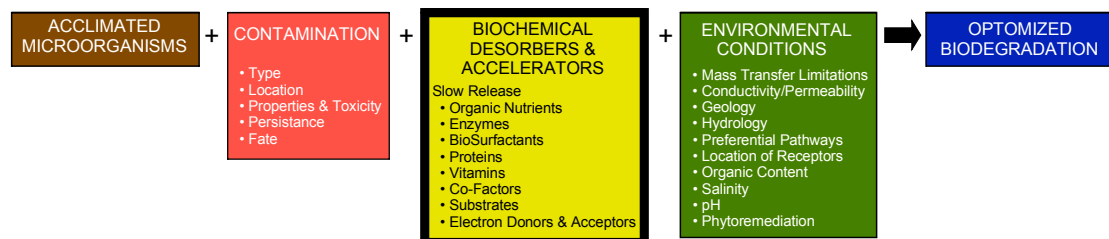


Figure 2: Acceleration of Bioremediation under Site Specific Conditions

2.3 Understanding and Overcoming Limiting Factors

Petroleum and chlorinated hydrocarbon biodegradation in soil and groundwater can be limited by many factors including nutrients, pH, temperature, moisture, oxygen, soil properties, contaminant type, and contaminant location. Biostimulation involves the modification of the environment with limiting additives to stimulate existing bacteria. This can be done by the addition of various forms of limiting nutrients and electron acceptors, such as phosphorus, nitrogen, oxygen, or supplemental carbon (Figure 2). The primary advantage of biostimulation is that bioremediation will be performed by native microorganisms that are present, distributed spatially, and adapted to the subsurface environment (5).

2.4 Overcoming Mass Transport Limitations

In order for bioremediation to take place, contaminants need to be transported to where microorganisms are most abundant, i.e., on fine soil particles. Since up to 60% of contaminants may be sorbed to soils, biosurfactants assist in the desorption and transfer contaminants from larger to smaller particles in the unsaturated and saturated zones where degraders are present (Figure 3).

Seventy-five to 90 percent of contaminant transport occurs within 10 to 20% of the aquifer cross section. Consequently, the velocity of contaminants is higher than previously predicted when passing through a limited area (1). Mass transport and uniform mixing are primary limiting factors to bioremediation. Soil and groundwater are generally heterogenous matrices that pose uniform mixing and transport challenges due to highly variable transport velocities and mass fluxes. Treatment zones need to be identified for technologies to come in contact or mix with contaminants and the environment to be effective (2).

Mass flux is defined by the mass per unit time per unit area that a contaminant plume is transported through a defined cross sectional area. Mass discharge is expressed as mass per unit time. Collecting mass flux information through three-dimensional data gathering techniques can provide information to better target contaminants in their respective zones. Tools for collecting this data include: electrical conductivity meters, pH probes, dissolved oxygen sensors, cone penetrometers, direct push lithology evaluations, oxidation/reduction potential, and passive flux meters (1).

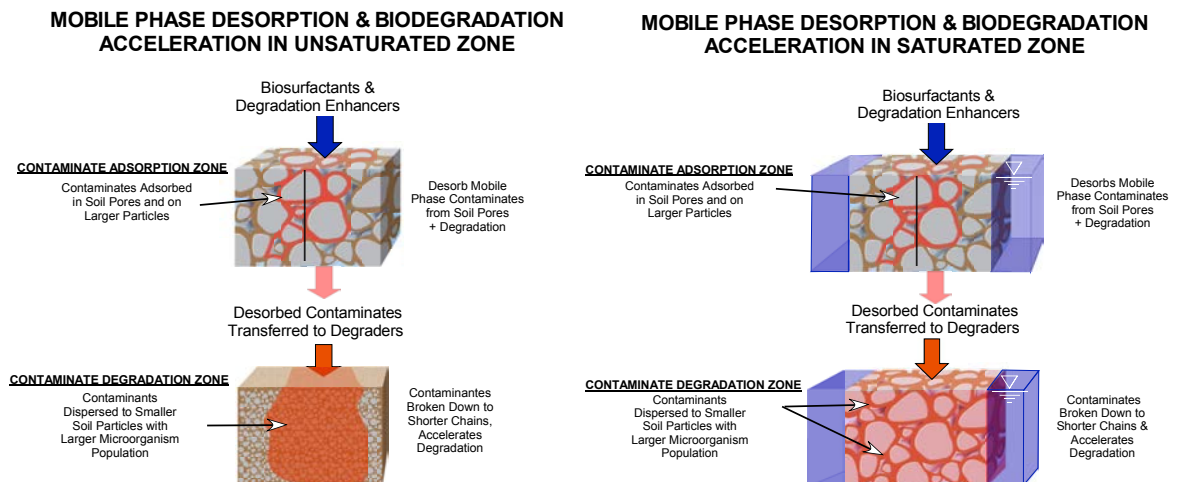


Figure 3: Mobile Phase Desorption & Degradation in the Unsaturated and Saturated Zones

2.5 Contaminant Bioavailability

Bound or adsorbed contaminants in soil pores restrict bioavailability (10). Bioavailability is a function of: the degree that contaminants are sorbed to solids or are sequestered by molecules in contaminated media, are diffused in macropores of soil or sediment, and other factors such as the low solubility of Light Non-Aqueous Phase Liquids (LNAPL) or Dense Non-Aqueous Phase Liquids (DNAPL). Bioavailability for microbial reactions is lower for contaminants that are more strongly sorbed to solids, enclosed in molecule matrices in contaminated media, are widely diffused in macropores of soil and sediments, or have low water solubilities (Figure 3)(5).

Microorganisms can produce reduced or oxidized species that cause metals to precipitate and be immobilized and tie up contaminants. Examples are: the oxidation of Fe^{2+} to Fe^{3+} , which precipitates as ferric hydroxide, reduction of SO_4^{2-} to sulfide, which precipitates with Fe^{2+} as pyrite, and reduction of hexavalent chromium to trivalent chromium. They can also degrade organic compounds that are bound to metals and keep metals in solution. Controlling pH, hardness, and alkalinity can determine the mobility of metals in the environment and bioavailability (11).

2.6 Contaminant Sequestration

Contaminant sequestration can limit the bioavailability of chemicals to degraders. Mechanisms involved in pollutant sequestration are (Figure 4):

1. Adsorption or precipitation on mineral surfaces
2. Adsorption on soft natural organic matter
3. Adsorption on condensed/hard natural organic matter
4. Diffusion in micro-porous media
5. Encapsulation

Bioavailability as it relates to organisms and contact with contaminants comprise several phase transition and mass transfer processes. Sequestration of contaminants can be due to one or a combination of the following factors:

- Physiochemical processes like adsorption, desorption and diffusion controlled by substance and specific soil parameters like hydrophobicity, aqueous solubility, pKa (index to express the acidity of weak acids), Cation Exchange Capacity (CEC), pH, clay and organic matter content.
- Physiological uptake processes (biological availability) controlled by species-specific parameters including anatomy, surface-to-volume ratios, feeding strategy, and habitats preferences.
- Toxicological availability controlled by organism metabolism, detoxification, storage capacity, excretion, and energy resources.

Diffusion kinetics into micro-voids of different pore size are non-linear. Tortuosity and interaction with pore walls limit diffusion, particularly in pores with diameters in the range of the contaminant's molecule size (12).

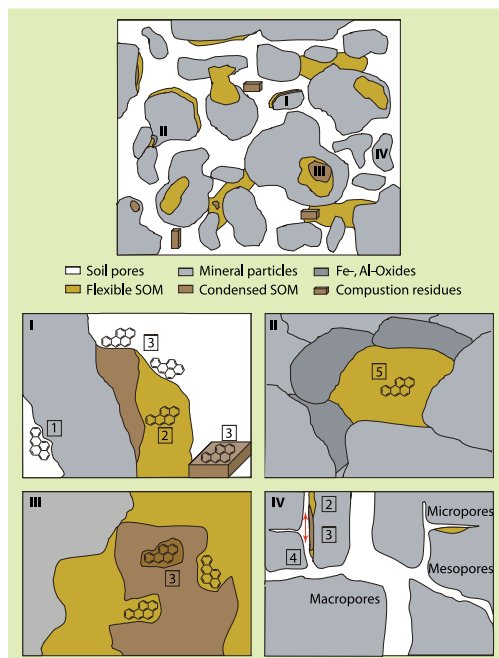


Figure 4 Top: schematic representation of potential sites for pollutant sequestration. Bottom: zoomed sections (varying scales): **I** – adsorption onto mineral surfaces and partitioning between water and organic phases; **II** – encapsulation of pollutants partitioned to soil organic matter; **III** – adsorption onto hard organic matter and capture of pollutants in hydrophobic cavities; **IV** – (retarded) diffusion of hydrophobic organic contaminants (HOC) in meso- and micro-pores (arrow). Numbers in rectangles refer to the sequestration mechanisms given in the list above. SOM = soil organic matter (12).

2.7 Particle Size Impact on Transport

Groundwater movement is determined by the immobile rock/soil phase, mobile water phase, and colloidal particles. Dissolved phase molecules or polymers are less than 0.1μ in diameter. Colloidal particles up to 2μ in diameter can move with groundwater (Figure 5) depending on soil porosity and particle size distribution. Clays, silt, sand, and gravel have different permeabilities that affect groundwater transport. Colloids are small organic or inorganic particles 0.1 to 10μ in diameter. Bacteria (0.5 to 5μ) may attach to colloidal particles. As this process progresses, biofilms may be formed that increase the size and mass of particles that leads to clogging of well screens and aquifers.

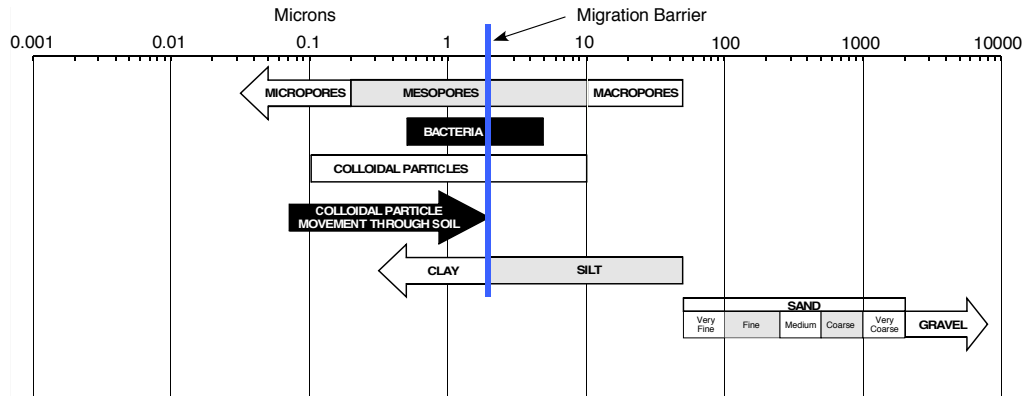


Figure 5: Relationship of Particle Size to Groundwater Transport

Soil characterization determines permeability and potential for uniform amendment addition. Clay soils are less permeable than sandy silt soils. Soil compositions of silt, sand, and gravel tend to have conductivities greater than 10^{-4} cm/sec that are more amenable to bioremediation. Soil investigations need to define variable compositions through the aquifer cross section and if less permeable barriers exist that could impede site communication. Short circuits caused by fractures or more permeable lenses can have a dramatic effect on contaminant transport. Backfill versus natural formations need to be identified since backfill can be random and prevent uniform transport.

2.8 Super Fine Powdered Activated Carbon & Nano Particle Amendments

Movement of colloidal particles through aquifers has increased interest in nano-particle amendments that include Super Fine Powdered Activated Carbon (SFPAC) and zero valent elemental metals for reductive dechlorination. These include elemental iron, tin, and zinc. SFPAC is of particular interest due to its ability to provide a particle host for biological growths to retain nutrients and to move through an aquifer and also provide chemical adsorption. Reduction in particle size dramatically increases the adsorption capacity of powdered activated carbon. This provides short-term benefits to soluble phase concentration reductions. SFPAC is generally not used as a source control, but for more dilute downgradient concentrations within a soluble contaminant plume. Long-term contaminant desorption, sorption exhaustion, biofilm buildup, and clogging aquifers are issues that need to be addressed as extended period monitoring data becomes available (13).

2.9 Biosorption & Biosurfactants

Petroleum hydrocarbons have a high sorption affinity with soil particulates, sediments, and organic content. Their water solubility is also relatively low. The functioning of microbial cells is to a large extent dependent on the aquatic environment. These compounds can also adsorb onto the soil matrix, which may result in a decrease in biodegradation efficiency (14). A compound's soil organic carbon partition coefficient (K_{oc}) is defined as the equilibrium condition between organic carbon and the contaminant concentration in an aqueous solution. Higher K_{oc} values indicate more contaminant mass is likely to be retained in soil and therefore less readily bioavailable. The opposite is true for lower K_{oc} values. Reviewing literature K_{oc} values can help predict the bioavailability of organic chemicals. The use of biosurfactants can assist in desorbing soil bound contaminants making them more bioavailable for degradation.

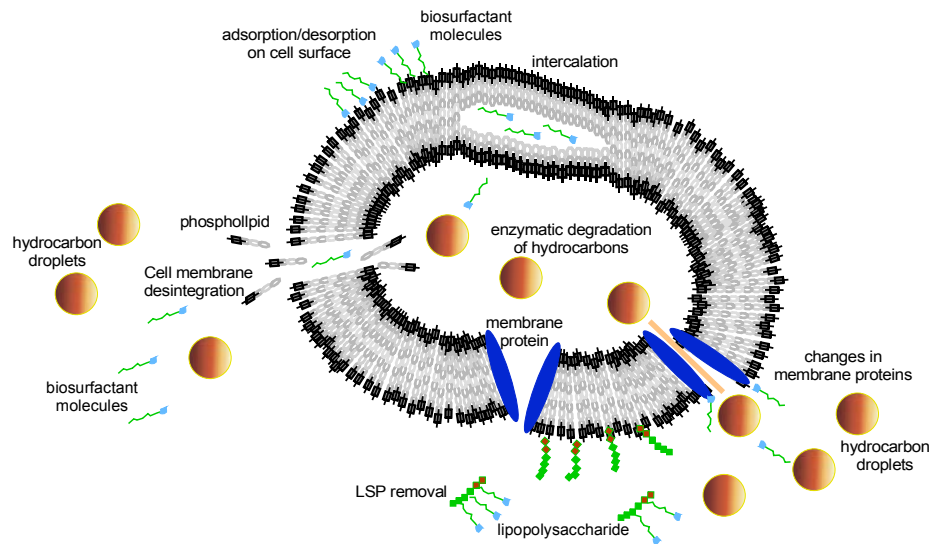


Figure 6: Interactions Between Bacteria Cells and BioSurfactants

Biosurfactants play an important role in bioremediation that include the solubilization and desorption of contaminants and modification of bacteria cell surface properties. Contaminant bioavailability is increased by solubilization and changes in cell surface hydrophobicity and membrane permeability. Hydrophobic contaminants are desorbed and emulsified and dispersed in the aqueous environment by reducing interfacial tension between immiscible liquids and increasing the solubility of hydrocarbons. Biosurfactants reduce the sequestration of pollutants by desorption followed by increased bioavailability of micronutrients.

Biosurfactants influence cell surface properties by altering cell surface hydrophobicity, electrokinetic potential, biomorphology, and surface functional groups. Cell surfaces are modified that promotes adhesion to hydrophobic contaminants or enhance their partitioning and bioavailability to microorganisms.

Biosurfactants increase the cells permeability or trans-membrane transport of contaminants which is the limiting factor in bioremediation (14) (Figure 6). Biosurfactants can also alter the cells fluidity that is a function of the lipid bilayer by altering the ratio of saturated to unsaturated fatty acids. Some researchers have hypothesized that biosurfactants are less likely to rupture cell membranes than synthetic surfactants. Biosurfactants impact on the cell can be summarized by decrease in cell surface hydrophobicity, loss of polysaccharides from the outer membrane, decrease in the cells electronegativity, aggregation, sedimentation promotion, and modifications of cells surface functional groups.

The action of biosurfactants leads to greater contaminant bioavailability and enhancement of microbial degradation. Natural biosurfactants are less toxic than synthetic surfactants and are more degradable than many synthetic surfactants. Carbon in surface active agents do not detract from available food source carbon in contamination that is the target of bioremediation.

2.10 Type and Location of Contamination

The biodegradability of organic constituents depends on their chemical structures and physicochemical properties, such as water solubility and water/octanol partition coefficient. Highly soluble low molecular weight compounds tend to degrade more rapidly while low water soluble complex compounds are less available and tend to degrade more slowly. Surfactants can enhance the degradation of more complex high molecular weight compounds by breaking them down into a more soluble phase (Figure 6).

Most low molecular weight contaminants (nine carbon atoms or less), aliphatic and monoaromatic compounds are more easily degraded than higher molecular weight aliphatic or polyaromatic organic compounds. Generally, the more highly chlorinated a contaminant, the more resistant it is to biodegradation. Weathered petroleum hydrocarbons are generally more difficult to degrade than fresh releases. Weathering and aging contribute to sequestration (4).

The fate of contaminants determines the location of reaction zones to deliver treatment amendments. DNAPL for instance, with densities greater than water, tend to migrate to lower depths than LNAPL. Contamination in the smear zone (between high and low groundwater tables) where LNAPL is frequently deposited, is less available for degradation. Knowing the location and type of contaminants is an integral consideration in targeting the location to deliver amendments to effective reactive degradation zones.

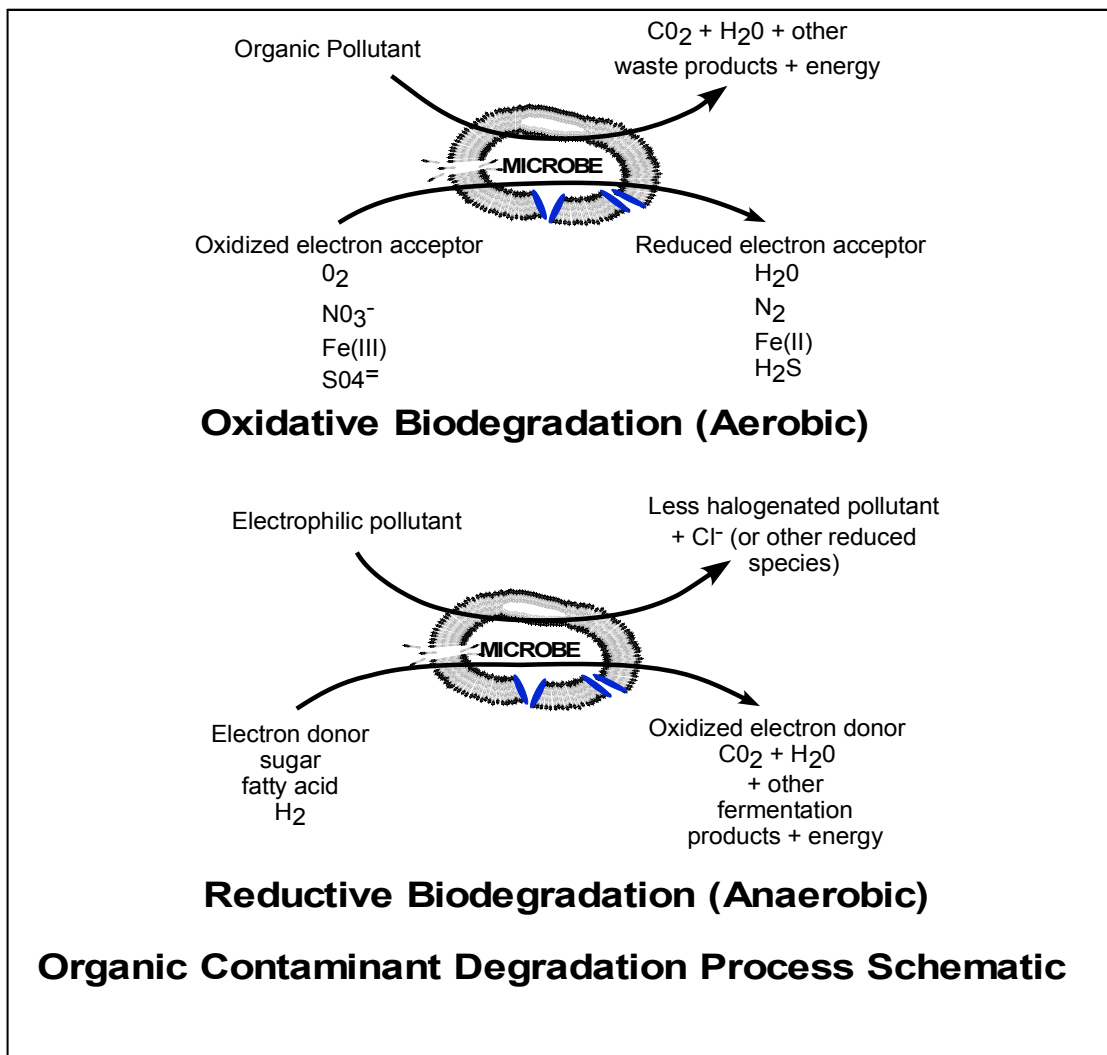
Accelerated aerobic degradation is frequently used in the vadose zone. With the rapid depletion of oxygen in areas of high contamination, anaerobic conditions rapidly develop, making anaerobic accelerants a de facto option. The location and soil properties contribute to the selection of the delivery method and type of amendment required (1).

2.11 Biochemical Catalysts

Biochemical transformations of pollutants require energy and catalysts. Microorganisms are the catalyst generator and enzymes are the catalysts. Enzymes cause catabolic reactions to provide energy and material for synthesis of additional microbial cells. Supplemental carbon may also be supplied for supporting cometabolic reactions. When biotransformation of contaminants leads directly to energy generation and growth of more microorganisms, the contaminant is known as a primary substrate.

Oxidation-reduction (redox) reactions are necessary for the generation of energy for microorganisms. Organic compounds are required to support microbial activity, i.e., an electron donor, contaminant or other organic compounds. Biochemical oxidization involves the transfer or release of electrons from contaminants to electron acceptors. Oxygen is an electron acceptor for aerobic reactions. Nitrates, manganese, iron, sulfates, and carbonate are the electron acceptor preference sequence for anaerobic reactions (Figure 7). Breakdown products of anaerobic degradation results in fermentation metabolism that produces acetate, propionate, ethanol, carbon dioxide, alcohols, organic acids, that can be degraded by other species of bacteria. Hydrogen is also produced which is a strong electron donor.

Figure 7: Organic Contaminant Degradation Process Schematic (21)



Oxidation-reduction (redox) reactions are necessary for the generation of energy for microorganisms. Redox potentials can assist in the following:

1. Identifying organisms that operate within a redox range.
2. Defining required electron acceptors or donors.
3. Predicting the predominant electron acceptors.
4. Predicting the stability of various compounds that regulate nutrients and metal availability.

Organic compounds are required to support microbial activity, i.e., an electron donor, contaminant or other organic compounds. Oxidizing systems accept electrons and reducing systems release electrons. The potential for a system to be aerobic or anaerobic can be estimated by redox potential. Lower redox potentials favor reducing conditions or anaerobic reactions while aerobic reactions with higher redox potentials favor aerobic reactions. Redox potential predicts the tendency of a reaction to

release or accept electrons and is measured in millivolts (mv). As depicted in Figure 8, as the redox potential of groundwater decreases, the predominant electron acceptor changes through the following sequence: oxygen, nitrate, manganese, iron, sulfate, and carbonate.

Aerobic reactions occur when oxygen is the electron acceptor. Organic overloading from petroleum releases consumes oxygen and rapidly creates reducing conditions. Aerobic organisms utilize oxygen as their terminal electron acceptor. For direct aerobic oxidation, primary reactants include oxygen and CAH (manmade organic compounds). Possible additives include air, oxygen, hydrogen peroxide (H₂O₂) and magnesium peroxide.

Facultative reactions involve organisms can use either oxygen or alternative electron acceptors.

Anaerobic reactions take place in the absence of oxygen where, nitrates, manganese, iron, sulfates, and carbonate are the electron acceptor preference sequence. Reducing conditions can cause specific metals to mobilize and create secondary water quality impacts. For anaerobic reductive dechlorination, primary reactants include hydrogen, organic carbon, carbon from a contaminant source (anthropogenic), and CAH (lactate, acetate, methanol, glucose, hydrogen, or molasses).

Breakdown products of anaerobic degradation results in fermentation metabolism that produces acetate, propionate, ethanol, carbon dioxide, alcohols, organic acids, that can be degraded by other species of bacteria. Hydrogen is also produced which is a strong electron donor.

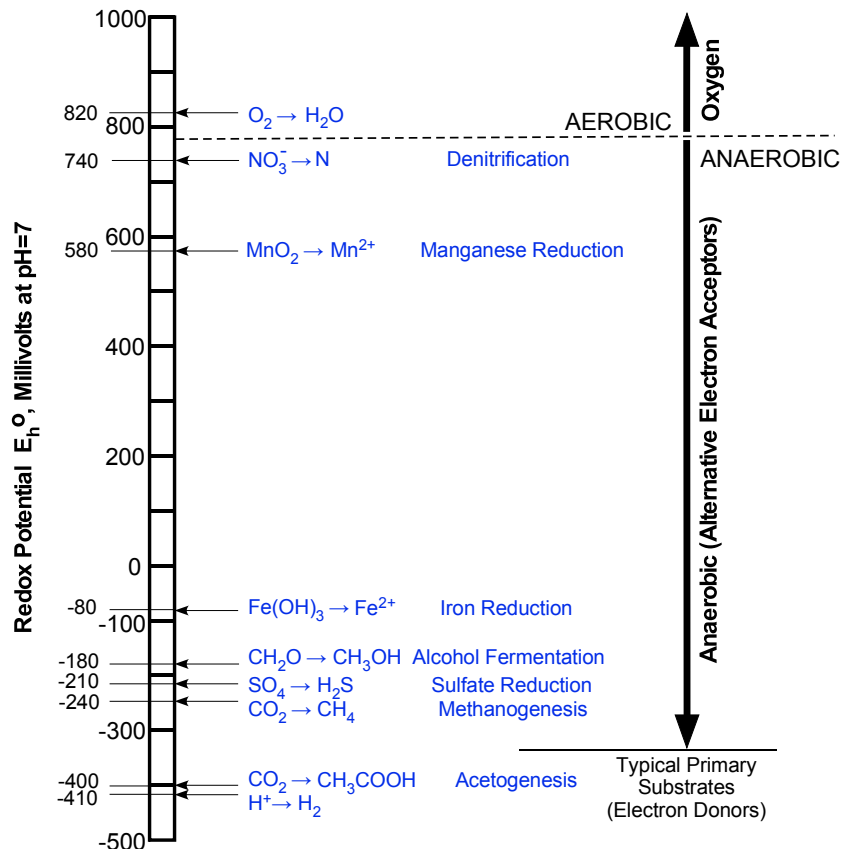


FIGURE 8: ESTIMATED REDOX POTENTIALS FOR VARIOUS ELECTRON ACCEPTORS (15)

Cometabolism is defined by a biotransformation when microorganisms use other carbon sources as their primary substrate rather than the contaminant to produce enzymes that degrade the contaminant. For cometabolic aerobic oxidation, additional primary reactants include organic or anthropogenic carbon and additional additives such as methane, propane, butane, and ammonia. Catalysts for cometabolism requires addition of a supplemental carbon source such as molasses. This is a particularly important metabolic pathway for anaerobic degradation of BTEX, PAHs, PCE, and TCE.

Abiotic transformations involve chemical transformations of contaminants without microbial degradation. Contaminants can be oxidized by oxygen additives such as hydrogen peroxide, ozone. Sulfide from anaerobic sulfate reduction can precipitate dissolved metals, including lead, cadmium, zinc, and copper. Zero-valent iron, supporting anaerobic organisms, produces hydrogen that abiotically reduces contaminants.

2.12 Dissolved Oxygen Forms

Aerobic respiration occurs when microbes utilize molecular oxygen as the electron acceptor to oxidize contamination. Anaerobic respiration occurs when oxygen is obtained from complexed oxygen in the form of nitrates, sulfates, or iron that serves as an electron acceptor. Oxygen can also be supplied by air, pure oxygen, hydrogen peroxide, ozone, or other oxygen releasing compounds (11).

The form of oxygen provided depends on site specific parameters. Injection of oxygen into reduced groundwater can result in rapid precipitation of iron, manganese, other metals, and biofilm within the well annulus and groundwater formation.

Air - Delivered as a gas by compressed air at 0.5 to 4.0 scfm. Oxygen concentration is limited to 8-10 ppm when the saturated zone is aerated using atmospheric air, which contains approximately 21% oxygen. Oxygen concentrations can be increased by above ground oxygen saturation and recirculation. The majority of projects are completed using this form of oxygen.

Pure Oxygen - Delivered as a gas. Dissolved oxygen concentrations of up to 40-50 ppm can be achieved through pure oxygen injection. There is an increased risk of fugitive vapor intrusion with the potential of fire or explosion. Relatively few petroleum hydrocarbon bioremediation projects have been completed with this form of oxygen.

Hydrogen Peroxide - Delivered as a liquid. Unstable and short-lived. Oxygen is wasted on oxidizing other organics. Relatively few petroleum hydrocarbon bioremediation projects have been completed using this form of oxygen. The precipitation of chemical oxidants (e.g., iron oxides) can present potentially significant equipment fouling problems. Potential dangers working with hydrogen peroxide should not be overlooked. Hydrogen peroxide is an oxidant that can cause chemical burns. When introduced at high concentrations, hydrogen peroxide can produce heat and elevated oxygen levels that may lead to fire or explosions by vapor intrusion. Using dilute concentrations of hydrogen peroxide can help reduce these hazards.

Ozone - Injected as a gas. Ozone is a strong oxidant with an oxidation potential greater than that of hydrogen peroxide. Ozone is 10 times more soluble in water than pure oxygen. Delivery is limited by mass transport and stability. Efficiency is limited because of its relatively low solubility and rapid decomposition in the aqueous phase. Some oxidized byproducts are not biodegradable. May have inhibitory effects on microbes. Relatively few petroleum hydrocarbon bioremediation projects have been completed using this form of oxygen.

Permanganate - Delivered as dissolved powder in water. Stable but is reactive with a narrow range of contaminants. May have inhibitory effects on microbes. Has a limited radius of influence.

Persulfate - Delivered as dissolved powder in water. Stable but requires activation for efficient degradation of contaminants. Activators include Fe^{2+} , Fe^{3+} , heat, H_2O_2 , and high pH. May have inhibitory effects on microbes. Has a limited radius of influence.

Calcium and Magnesium Peroxide - Delivered as a solid. Lasts for 9 to 18 months. May have inhibitory effects on microbes. Releases oxygen for four to eight months. Ten percent is delivered as oxygen. Has a limited radius of influence.

Mechanical aeration is generally less expensive than peroxide, ozone, or chemical oxygen supplements (1). Aerobic degradation of petroleum hydrocarbons and other contaminants can rapidly deplete dissolved oxygen concentrations in soil and groundwater. Subsequently, anaerobic bioremediation becomes an important metabolic pathway. Aerobic oxidization can also cause biofilms to form at amendment injection points.

Biofilms can be minimized by alternating between aerobic and anaerobic degradation states or adding enhancers in a pulsed fashion. A water chase following accelerant additions can also minimize the formation of biofilms. Biofilms can be removed by well rehabilitation using hydrogen peroxide or carbon dioxide injected under pressure.

2.13 Contaminant Overloading

Contaminant concentrations directly influence microbial activity. When concentrations are too high, contaminants may have toxic effects on native bacteria. In contrast, low contaminant concentrations may prevent induction of bacterial degradation enzymes (5). For bioremediation to be effective, free product or saturated petroleum matrices need to be removed. Soil TPH concentrations greater than 50,000 ppm and solvent concentrations greater than 7,000 ppm can retard biochemical reactions (6).

Biosurfactants or commercial surfactants can be used to accelerate the removal of free product or sorbed contaminants by soil washing prior to initiating enhanced bioremediation. Biosurfactants are preferred over synthetic surfactants due to their lower toxicity. Biosurfactants are also produced by native microbes during the degradation process. The higher the initial TPH concentrations on soil or ballast the longer it takes to reduce concentrations to acceptable levels. This is reflected in Remtech's data on degradation of petroleum hydrocarbons on track siding case histories 8, 10, and 11 presented later in this paper.

2.14 Groundwater Velocity & Contaminant Mobility

Contaminants move through soil and groundwater according to matrix permeability, soil particle size, hydraulic conductivity, advection, dispersion, diffusion, hydraulic gradients, and capillary action. The dominant factors are advection and diffusion (1).

Reaction kinetics of microbial degradation requires adequate retention time of biochemical enhancers. Slow release and/or increased density and frequency of amendment applications can provide sufficient quantities prior to being washed away. Recirculation of amendments can also assist in maintaining supplemental concentrations.

In groundwater applications, hydraulic conductivities greater than 10^{-4} cm/sec are desirable for enhanced bioremediation, although lower conductivities 10^{-4} to 10^{-6} cm/sec may be technically feasible, but costlier. Hydraulic conductivity can be quite variable across a site. When the hydraulic conductivity is determined by slug tests at multiple locations and depths, more accurate velocities can be calculated. Groundwater velocity is impacted by hydraulic gradient, hydraulic conductivity and porosity or according to Darcy's equation.

Darcy's velocity is expressed as:

$$V_D = -K_H (\Delta h / \Delta X)$$

K_H = hydraulic conductivity
 $\Delta h / \Delta X$ = hydraulic gradient

2.15 pH and Biofilms

Most aerobic bacteria grow in the pH range of 5 to 9 whereas anaerobic bacteria thrive in the range of 6 to 8. Some reductive dechlorination can occur at pHs less than 5 but may increase the solubility of many metals and metalloids. The optimal pH is site specific. For example, vigorous microbial activity is present in some natural systems with pHs of 4.5 to 5. Adjusting pH to neutral conditions can inhibit the microbial activity of some organisms (6).

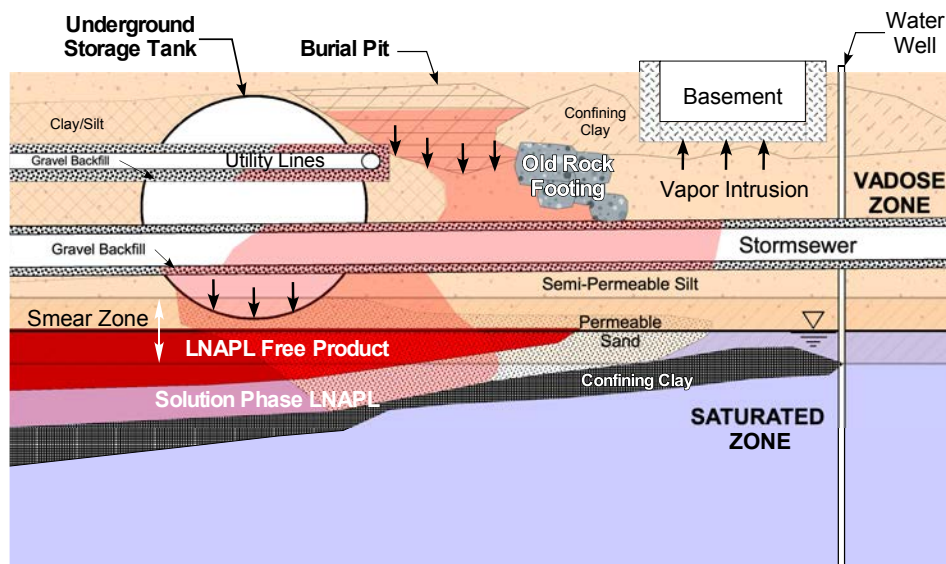
When pH adjustment is necessary, acidic pH buffering can be achieved by the addition of soda ash, lime, calcium hydroxide or sodium hydroxide in order to raise the pH. When basic buffering is required, citric acid, hydrochloric acid or other acids can be added to lower the pH. Changes in pH should be no more than 1 or 2 units, as rapid changes can shock or retard biological activity. Raising the pH too much can result in mineral precipitation and clogging. Hardness above 400-500 mg/l (carbonate) in receiving waters can lead to the precipitation of heavy metals. Lowering the pH may cause metals to mobilize. Acid or base titration tests can be useful in determining acceptable pH changes due to amendment additions (6).

Injection of substrates and nutrients enhances the growth of microorganisms that exist as suspended flocs or surface biofilms. Biofilms form when large aggregations of bacteria and microbes bind together in an extracellular polymeric structure that attaches to particle surfaces. This polymeric structure consists of polysaccharides, nucleic acids, and proteins that form a protective armor which insulates microorganisms from geochemical changes.

Biofilm growths are more significant under aerobic rather than anaerobic conditions. Operating systems in an intermittent mode between aerobic and anaerobic conditions can limit biofilm growth and clogging around injection points and in the aquifer. Increasing intervals between injection events followed by a clean water chase can limit biofilm formation. Temporarily lowering the pH can also minimize the formation of metal precipitates, but solubilizes and mobilizes metals that need to be controlled. Lowering the pH below 5.5 can impact the activities of nitrate reducers, sulfate reducers, iron reducers, and methanogens.

Biofilms also often require well rehabilitation employing chemicals such as hydrochloric acid, hypochlorite, hydrogen peroxide, and glycolic acid. Mechanical methods include jetting, surge blocking, over pumping, and brushing.

FIGURE 9: SAMPLE PREFERENTIAL CONTAMINATE PATHWAYS & SENSITIVE RECEPTORS



2.16 Contaminant Migratory Pathways

Locating contamination in soil and groundwater can be difficult (Figure 9). In one author's experience, it can be like trying to find a *needle in a haystack*. Contaminant concentrations in various pathways can vary several orders of magnitude due to manmade or natural features. Natural features include variable permeabilities, variable soil compositions, confining layers, formation fractures, sorbability of contaminants, perched groundwater tables, or contaminants located in the smear zone.

Examples of manmade preferential pathways include permeable backfill around leaking underground storage tanks, utility lines, old foundations, waste burial pits, backfill areas, septic tank drainage fields, dug or drilled water wells, and sanitary sewers or stormwater lines.

Frequently, a difference of a couple of feet horizontally or vertically can make it difficult to locate and target contaminants. This happens regardless of predictive attempts to locate pathways from records, utility locations, geophysical surveys (such as ground penetrating radar), soil sample logging, tracer tests, and other testing techniques. Invariably, there is one sample location or a remaining *hot spot* that requires further remediation to achieve cleanup limits.

For example, several feet of gasoline were discovered at a construction site of an 18 million dollar building in a downtown area during elevator shaft rock blasting operations. The state fire Marshall cancelled the construction permit. Consultants initially engaged predicted that there were thousands of gallons of gasoline in fractured rock under the site due to several feet of fuel observed in several monitoring wells. This large volume of fuel was questionable due to the fact that there was no record of fuel pipelines in the area.

To reinstate construction permits, the fire Marshall required that the source be found, removed, and that soil vapors be reduced to less than 5% of the Lower Explosion Limit (LEL). An old Sandborne map was discovered in the attic of the City Manager's office that placed an UST under the contractor's construction trailer. Exploratory excavations uncovered remnants of UST that had been removed several decades earlier. Tracing the gasoline finger print down a 20-foot open excavation confirmed the pathway of free product. Test pumping demonstrated that gasoline in rock fractures was connected and could be recovered by pumping. Less than 100 gallons of gasoline were removed and flammable vapors in test locations met the Fire Marshal's requirements. Building permits were reinstated and the building was completed. As a precautionary measure, a ventilation and recovery well system was built into the building's foundation. Measurements following construction completion revealed that no free product or gasoline vapors remained.

2.17 Selection and Toxicity of Amendments

Amendment Selection

To select effective amendments the following considerations should be addressed:

- Are amendments appropriate for the contaminants and the targeted reactive zones?
- Are sufficient concentrations and retention times maintained to degrade contaminants?
- Are amendments or secondary reactants migrating outside reaction zones?
- Should hydraulic capture, containment barriers, or recirculation be considered?
- Are contaminants causing concentration overload to microbes?
- Are nutrient addition overloads being prevented?
- Are metals mobilization or precipitation, biofilm formation, or unintended impacts controlled?
- Are amendments soluble and moving freely through the reaction zone?
- Are particulates in amendments restricting movement through the aquifer?
- When degradation is complete, are amendment toxicities reduced to non-toxic levels?

The appropriate biostimulation agent(s) should be selected for the appropriate zone and may contain nutrients, electron donors, electron acceptors, supplemental carbon sources, or oxygen.

- Nutrients - All Zones
- Anaerobic Respiration Agents - Anaerobic Zone
- Oxidizing/Aerobic Compounds - Aerobic Zone
- Cometary Oxidization Amendments - Aerobic Zone
- Anaerobic/Dechlorination - Anaerobic Zone

Amendment Toxicity

The toxicity and efficacy of amendments should be considered. Aquatic fish 96 hr LC₅₀s toxicities greater than 5,000 mg/l are considered relatively non-toxic and less than 100 mg/l are considered acutely toxic. A significant number of amendments are acutely toxic to aquatic organisms (Table 1). Some amendments are more toxic than the petroleum hydrocarbons they are intended to degrade. In addition, intermediate products of degradation can be more toxic than the contaminants that are being degraded. When selecting amendments it is important to check the safety data sheets and confirm human and environmental toxicity values to protect public health and the environment.

Foreign Microbes, included in some amendments, are prohibited in several states due to their potential to form toxic byproducts, toxicity to native microbes, or interference/competition with the complex native microbial degradation populations.

Several controversial and unknown issues are associated with the addition of foreign microbes:

- 1) How does competition with native microbes impact overall degradation outcomes and toxicity?
- 2) When foreign microbes are introduced, how do you distinguish between the degradation of introduced microbes and natives?
- 3) Not all native degraders have been identified and may be present below present detection limits or analytical discovery methods.
- 4) Why do bench-scale tests that indicate select or genetically engineered microbes will degrade select contaminants in lab but sometimes die in the field?
- 5) Bacteria size limitations may prevent distribution into treatment zones (See Figure 5)
- 6) Ideal biostimulation environments should be established prior to introducing foreign microbes
(2) — How does altering conditions to foreign microbes impact other native microbes in other zones which have their own preferential conditions for degradation?
- 7) Can supplements and foreign microbial populations be supplied in a manner that is sustainable through the reaction period?
- 8) Will native populations be restored following completion of augmentation?

The use of native populations that are already mixed in the environment, the majority of which can degrade petroleum hydrocarbons, and small non-detectable concentrations of degradable microbes can be proliferated through biostimulation, is why in most cases biostimulation of natives is the preferred approach (1, 6).

Nutrient Requirements and Toxicities - Microorganism nutrient requirements are approximately similar to their cell composition. Nitrogen and phosphorous overloads can cause oxygen depletion, biofilms, algae and macrophyte growths, and toxicity to microbes. Nutrient overloads can be avoided by maintaining the proper carbon to nitrogen to phosphorous to potassium (C:N:P:K) ratios. Typical C:N:P ratios 100:5:1 may be required, with a considerable amount of the carbon supplied by contaminants. Others have determined the C:N:P ratio to degrade PAHs is C:4.67:1. When potassium is considered, the C:N:P:K ratio is 100:5:1:1 (16). For PAH degradation, phosphorous may be the dominant macronutrient. Micronutrient requirements for sulfur to calcium to magnesium (S:Ca:Mg) ratio is 2:1:1. Actual nutrient requirements can vary widely depending on the contaminant, microbial population, and degradation zone (1, 17).

Organic nutrients are less toxic than inorganic nutrients (Table 1). Only two listed organic nutrients, HC-2000 and urea, have aquatic toxicities greater than 5,000 mg/l. Fifty percent of the nutrients listed in Table 1 are acutely toxic and more toxic than gasoline and E85 gasoline. Twenty-nine percent are more toxic than diesel fuel.

2019 NCP Bioremediation Product Toxicities - Only two of the listed NCP products have aquatic toxicities greater than 5,000 mg/l with one of the products having foreign microbes. 77.4% are acutely toxic or contain foreign microbes. 29% are more toxic than gasoline or E85 gasoline. 22.6% are more toxic than diesel fuel. As mentioned previously, foreign microbes injected into the environment are prohibited in several states due to their potential to form toxic byproducts or interfere with the complex native microbial population.

NCP bioremediation products are specifically tested by the EPA for use in saline or ocean environments to degrade crude oil and not freshwater or refined products. Foreign microbes cultured and selected to degrade crude oil introduced into a beaker don't have to compete with a host of native microbes. This presents a potential bias towards foreign microbes when native microbial populations are, in most cases, considered the primary degraders. This potential bias seems to be indicated with nine of the fourteen NCP products with the highest alkane and aromatic degradation efficiencies, alkanes >90 percent and aromatics >67 percent, containing foreign microbes.

Different efficacy testing methods are required for the biostimulation of inland freshwater environments when stimulating native bacteria. Degradation efficiencies are also different for refined petroleum products versus crude oil. EPA testing protocols do not allow for the quantification of the acceleration of native microorganisms.

Anaerobic Respiration Compound Toxicities - Only HC-2000 of the listed amendments is relatively non-toxic and 50 percent are acutely toxic and more toxic than gasoline and E85 gasoline.

Anaerobic/Dechlorination Agent Toxicities - Only HC-2000 of the listed amendments is relatively non-toxic. Fifty-eight percent are acutely toxic and more toxic than gasoline or E85 gasoline.

Aerobic Oxidizing Additive Toxicities - None of the listed supplements are considered relatively non-toxic. 62.5% are acutely toxic and more toxic than gasoline and E85 gasoline.

Cometabolic Oxidization Agent Toxicities - Only HC-2000 is considered relatively non-toxic. 75% of the listed additives are acutely toxic and more toxic than gasoline and E85 gasoline.

Buffering Compound Toxicities - None of the listed buffering compounds are considered relatively non-toxic. 57.1% are considered acutely toxic.

2.18 Bioremediation and Phytoremediation Synergies

Petroleum based contamination located in wetlands, swamps, creek banks, ponds, detention basins, drainage ditches, riparian buffers, or heavily vegetated areas are candidates for phytoremediation that can enhance the bioremediation process. Phytoremediation strategies are particularly applicable to sites with shallow groundwater tables and are within the influence of plant root zones.

Plants and wetland soils, in many cases, are an ideal location to accelerate the degradation of select contaminants. Vegetated wetland soils can have microbial populations ranging from 2×10^3 to 5×10^6 CFU/gm (bacteria, actinomycetes, and fungi). In areas with shallow groundwater tables or where hydric soils are present, petroleum hydrocarbons, BTEX (See Case Histories 1 and 7), PAHs, and chlorinated solvents have been effectively degraded. The hydraulic conductivity of sedge, reed, and alder wetlands range from 1.16×10^{-2} cm/sec to 1.16×10^{-4} cm/sec when saturated (4). Wetland soils are within the desired range of conductivities for potential bioremediation.

Table 1: Amendment Comparative Aquatic Fish Toxicities (96 hr LC₅₀), mg/l

Comparative Amendment Fish Aquatic Toxicities (96 hr LC₅₀)

Petroleum Hydrocarbons	Anaerobic Respiration	2019 NCP Bioremediation Products
1,2,4, Trimethylbenzene 7.19 - 8.28	Ammonium Nitrate 100	Act Terra Firma (Act-tf) 2.61 FM
Benzene 5.3	Carbon Dioxide 35	Agroremed (Spill Remed) 37.5 FM
Diesel Fuel 35 - 276	Ferrous Sulfate 42	B&S Industrial (step one) Surface
E85 Gasoline 100	Potassium Nitrate 1,378	Bet Biopetro FM
Ethylbenzene 4.0	Sodium Nitrate 1,107	Bio-Regen Hydrocarbon FM
Fuel Oil, No. 2 35,36	Sodium Persulfate 771	BioRem-2000 Oil Digester FM
Fuel Oil, No. 2 35,36	Sodium Thiosulfate 28.5 to 88.3	Biosolve 7.4
Fuel Oil, No. 6 3.1	HC-2000 5785	Bioworld BioTreat Products 13.9 FM
Gasoline 82-119		Drylet Mb Bio FM
Hydrogen sulfide 0.002	Oxidizing/Aerobic	Dualzorb (Whitzorb) 8.43 FM
MTBE 773	Calcium Hydroxide 33.88	Ergofit Micromix Aqua FM
Naphthalene (Kerosene) 18 - 25	Hydrogen Peroxide 142.8	F-500 <10
Toluene 5.8	Oxygen ND	Hydroremed (Spillremed) 37.5 FM
Xylenes 4.2 - 8	Ozone 0.0093	JE1058BS 8.64 FM
	Sodium Persulfate 771	Land & Sea Restoration 001 FM
Nutrients	Sodium Permanganate 3.6 - 4.6	Micro-Blaze 4
Ammonium Nitrate 100	Potassium Permanganate 3.6	Munox SR FM
Ammonium Molybdate Tetrahydrate 320-420	Persulfate 76.3	Oil Spill Eater II (OSE II) 5258
Boric Acid 1,021	Magnesium Oxide 428	Oilclean (Proact) FM
Calcium Chloride 4,630		Oppenheimer Formula 16000 FM
Calcium H2 Othophosphate >13.5	Dechlorination/ Anaerobic Compounds	Petroclean 115 FM
Diammonium Phosphate 1700	Butyrate (48 hr) 53	Remediade Nutrients
Dipotassium Phosphate >100	Citric Acid 625	S-200C 25.33
Disodium H2 Phosphate Dihydrate 467	Dipotassium Phosphate >100	Shamantra Green Nutrients
Iron(III) Chloride Hexahydrate 22	Ethanol 375.00	Simple Green 8.3
Magnesium Sulfate Heptahydrate 15.5	Ferrous Sulfate 42	Soil RX FM
Manganese Sulfate Monohydrate 14.5	Glycerin 50-67	Sump Safe Bio-Reclaim FM
Monopotassium Phosphate 80 to 105	Glycerol Tripolylactate 51 - 57	System E. T. 20 FM
Potassium Nitrate 1,378	HC-2000 5,784	VB591, VB997, Binutrix Nutrients
Sodium Nitrate 1,107	Iron 0.63 - 1.99	Waste Away 12.54
Sodium Phosphate, Tribasic 28.5 to 88.3	Iron Sulfide >10	WMI-2000 FM
Sodium Thiosulfate, 72 hr 178	Lactic Acid 257.73	
Tricalcium Phosphate >2.14	Methanol 8	Buffering Compounds
Urea 6,810	Molasses ND	Sodium Hydroxide 33 - 189
Zinc Sulfate Heptahydrate 12.2	Monopotassium Phosphate 80 to 105	Calcium Hydroxide 33.88
HC-2000 5,784	Phosphorous Compounds >100	Citric Acid 440 - 760
	Propionate 39 - 77	Hydrochloric Acid 282
Cometabolic Oxidization	Sodium Benzoate 484	Phosphoric Acid 88.3
Butane 27.98	Sodium Nitrate 1000	Sodium Carbonate 300
Methane 10 - 100	Sugars ND	Sulfuric Acid 42 - 60
Propane 27.98	Vegetable Oil ND	
HC-2000 5,784	Zinc Oxide 0.46 - 1.06	

Notes: Aquatic Toxicities from EPA, SDS, and other data bases. Toxicities may vary for different species and data sources. FM = foreign microbes, ND = No Data

Degradation in soil around the root zone of plants is referred to as rhizodegradation. Plant degradation mechanisms are: phytosequestration, rhizodegradation, photohydraulics, phytoextraction, phytodegradation, and phytovolatilization (1, 4).

Rhizodegradation results from soil microbial degradation activity in the rhizosphere (root) zone that includes yeast, fungi, and bacteria. Plant roots release sugars, alcohols, and acids that contain organic carbons that serve as nutrients to degrade fuels, petroleum products, PAHs, chlorinated solvents, and munitions. Plant roots also loosen/aerate soil and introduce oxygen and water. Microbial concentrations in rhizosphere zone can be twenty times greater than in non-rhizosphere zones and degrade contaminants faster than in unvegetated areas. Nutrient, biosurfactant, and enzyme additions can accelerate degradation.

Phytosequestration in the rhizosphere occurs when plants absorb contaminants in roots or adsorb on roots and effectively immobilize or reduce the bioavailability of contaminants. Contaminant sorption can occur on both soils and plant roots.

Photohydraulics involves the uptake and transpiration of water by plant roots. In areas with shallow groundwater tables, contaminant containment (or hydraulic barriers) can be achieved by depressing the groundwater table where deep rooted trees are present. This applies to water soluble and leachable contaminants that are not phototoxic to plants. Roots also penetrate the sorbed contaminant zone and soil micropores and mobilize bound contaminants for degradation. Photohydraulic mechanisms attenuate and contain BTEX, chlorinated solvents, PCBs, pesticides, and inorganic contaminants.

Phytoextraction transfers contaminants into plant roots, shoots, trunks, and leaves. Contaminants are stored, sequestered, metabolized, or phytovolatilized from the transpiration flow leaving the plant. This mechanism is effective on contaminants such as PCBs, and inorganics (As, Cd, Cr, Cu, Ni, Se, radionuclides). Metals are generally removed by plant harvesting, composting, incinerating or landfilling.

Phytodegradation degrades contaminants within the plant by plant produced enzymes. Plant roots are not discriminating in the uptake of small organic molecules. Soluble molecules pass through the root epidermis and translocate through the plant. Less soluble compounds may be adsorbed or accumulated in plant tissues. Many contaminants are bound on the root surface and are not translocated. Potential contaminants for plant uptake have K_{ow} s < 3.5 and > 1.0 . K_{ow} s less than 1.0 are not actively transported through plant membranes. Contaminants such as BTEX, chlorinated solvents, short-chained aliphatic chemicals, aromatic amines, phenols, pesticides, herbicides, have K_{ow} s in the range for plant translocation for destruction of contaminants.

Phytovolatilization occurs when plants uptake, translocate, and release volatile compounds to the atmosphere through their leaves. Phytovolatilization works on volatile organic compounds, herbicides, and pesticides (1).

2.19 Injection Permits & Hydraulic Controls

Groundwater injection of amendments may require underground injection permits. The use of food quality supplements are generally preferred by most regulatory authorities. Secondary groundwater or drinking water standard exemptions may be required if suspended solids, pH, total dissolved solids, or COD concentrations are temporarily exceeded. Hydraulic controls may also be required, especially if downgradient sensitive receptors are present or to ensure that plume containment is maintained.

3. BIOSTIMULATION DELIVERY METHODS & REGULATORY ENVIRONMENT

Certain restrictions are placed on the selection of technologies when cleanups go through regulated or reimbursement programs. The command and control regulatory structure, along with institutional rigidity, may have become increasingly out of step with the demands and realities of environmental protection. Realization that returning a site to pre-contamination conditions or drinking water standards with existing technologies is not possible for certain contaminated sites under complex geologic conditions. This approach frequently is contrary with implementing innovative and cutting-edge technologies. Solutions require exchange and education of researchers and practitioners in the private and regulatory communities to select and implement best available and cost-effective remediation technologies. What is required is a multidisciplinary background in the sciences and engineering to understand and implement these complex issues (1). Considerable flexibility is frequently offered when a remedial project is taken through a voluntary cleanup program with regulatory approval.

In one of the author's experience on over 6,000 remediation projects since 1975, responding to spills, that serve as pilot tests, and lessons learned on what works, why, and why not, has been useful in progressing from small to successful large-scale remediation projects.

Understanding the "art of delivering the tool" is frequently more important than the tool. Many bioremediation supplements are proprietary and ingredients are not known to general practitioners. Data bases are not always known by the manufacturer and are frequently distributed between many users making it difficult to evaluate and select successful amendments.

Various **predictive** tests can be useful tools in defining treatment zones. Predictive test methods include; vacuum, pressure, dye tracer, draw down, test borings, monitoring wells, and direct-push sampling. In fractured rock, for instance, drawn down pump tests are an effective way in determining if fractures are interconnected and treatment is possible.

In one of the author's experience, the sooner site connectivity, or lack thereof, can be **demonstrated**, the sooner a successful remediation program can be implemented. Nevertheless, full-scale remediation programs generally require adjustments to delivery systems to establish communication with the entire site.

Amendment quantities can be estimated based on stoichiometry, estimates of biological demand, rules of thumb, and the results of bench scale or pilot tests. Consistent amendment concentrations at a uniform rate generally produce optimal treatment efficiency and limit secondary geochemical effects (18). Amendment concentrations, rates of delivery, and quantities need to be field adjusted due to competing mass transport conditions, amendment sorption, potential precipitation, potential biofilm formation, competitive microbial utilization, and variable delivery/diffusion through different reactive zones (2).

3.1 Delivery Methods

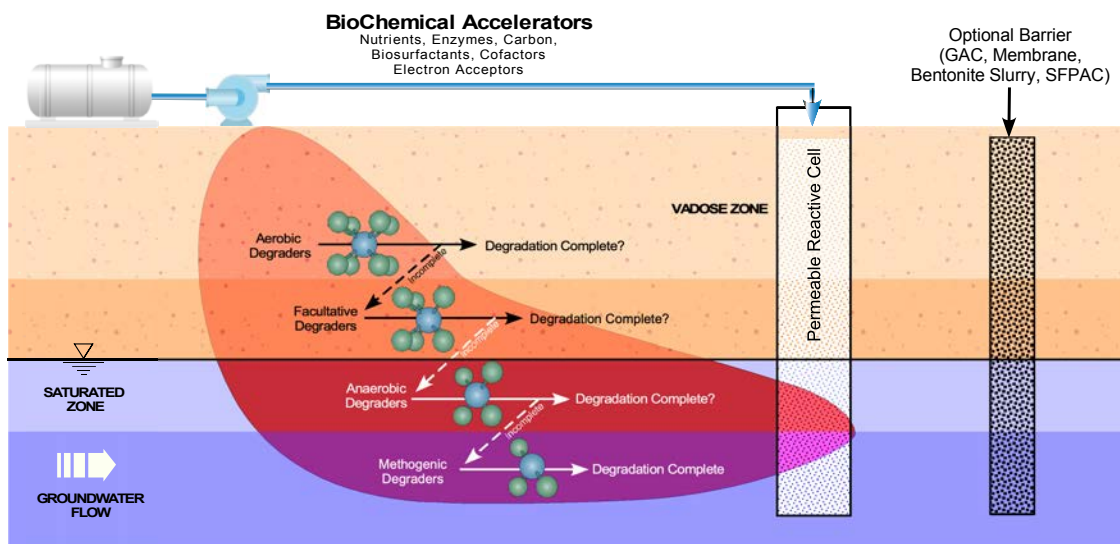
Delivery methods may be classified as active, intermittent, and/or passive. The spacing and depth of injection points requires designing an appropriate grid that provides uniform application to the soil and groundwater matrix where contamination is located.

When delivering amendments, consideration has to be given to hydraulic capture and the toxicity of amendment leachates to downgradient receptors. This is particularly important if degradation kinetics are slower than the retention time required for complete mineralization. Increased density of treatment grids, controlled dosing, or barriers are other methods that can control amendment transport and reaction kinetics.

3.1.1 Passive Delivery Systems (Figure 10)

Passive systems generally work in permeable formations with adequate groundwater gradients. Surface spraying of amendments can treat shallow vadose zones. Surface or deep tilling can increase permeability and enhance the vertical migration of surface applications. Permeable injection cells constructed with geotechnical fabric and filled with gravel can be charged with amendments in order to enhance percolation. Large diameter horizontal screens with vertical risers can also be used to feed supplements. Optional barriers constructed out of membranes, bentonite slurry walls, or sorbents can be used to attenuate downgradient transport.

FIGURE 10: SAMPLE PASSIVE PERMEABLE REACTIVE BARRIER INJECTION SYSTEM



3.1.2 Active Delivery Systems (Figures 11, 12)

Active delivery systems consist of pressure injection of accelerants through vertical and/or horizontal well screens. Injection wells should be installed with appropriate screen lengths, depths, and properly sealed to prevent short circuiting between well casings and boreholes.

A cement grout is generally required to ensure adequate sealing. Injection pressures should be as low as possible to prevent rupturing of wells. Screens need to be adequately sized to deliver the design flowrate while pressurizing the entire section to ensure appropriate mass transfer to the intended subsurface region. Variable depth injection points may be required through different permeability zones to ensure contact with contaminant locations.

Active delivery systems can be designed with or without hydraulic control or recirculation systems. Active systems without hydraulic control may consist of reversible systems where fluids are injected or withdrawn from the saturated zone. Hydraulic controls may be required in tighter soil formations where biosurfactant soil washing needs to occur to remove sorbed or mobile phase contaminants. Barriers as described in passive systems, may also be utilized. Sparge points may also be used in the saturated zone (Figures 11, 12).

Draw down and recirculation can increase flowrates, contact, and stabilize amendment concentrations. Hydraulic control can also ensure that solution phase contaminants and amendments do not migrate outside the treatment zone.

3.1.3 Intermittent Delivery Systems

Intermittent systems may alternate between active and passive modes. Pulsed applications may be effective to minimize over-oxygenating groundwater and subsequent biofilm growth on injection points or in the aquifer. Sufficient time may be required between supplement applications to allow adequate reaction time. Raising and lowering groundwater tables can also assist in removing and treating the

FIGURE 11: SAMPLE NON-HYDRAULICALLY CONTROLLED ACTIVE INJECTION SYSTEM

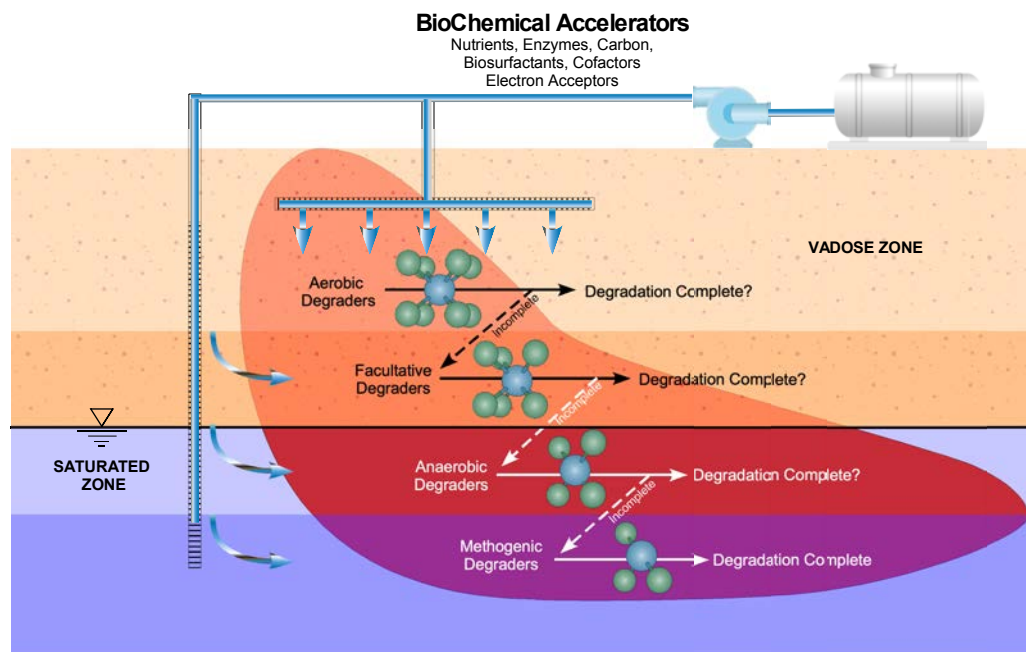
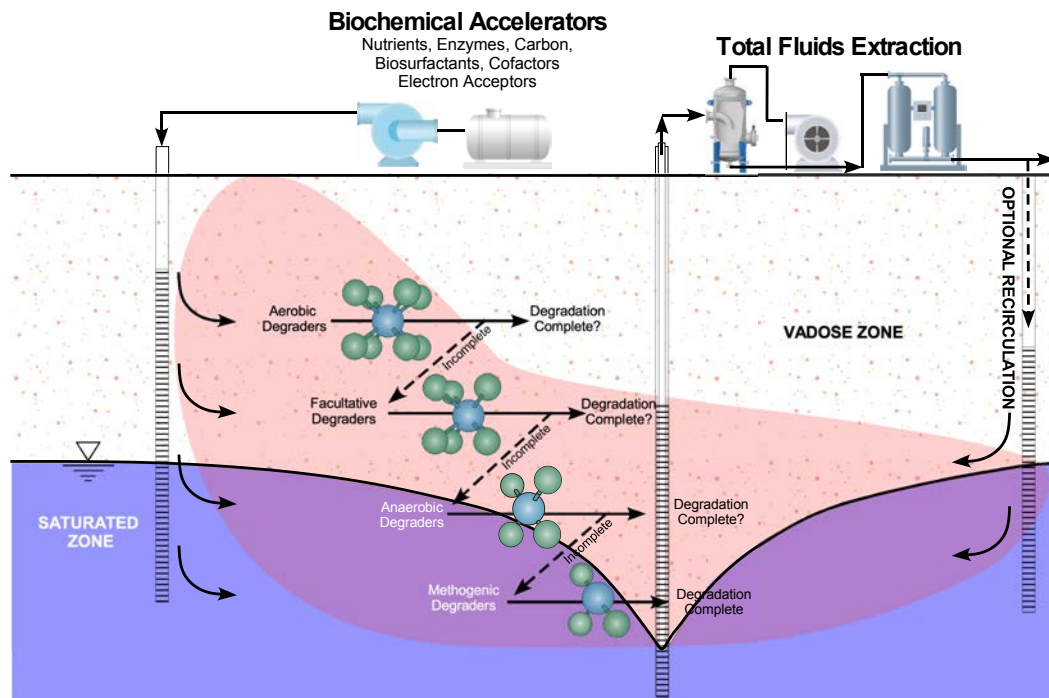


FIGURE 12: SAMPLE HYDRAULICALLY CONTROLLED ACTIVE INJECTION SYSTEM



4. OPTIMIZING BIOREMEDIATION WITH HC-2000

4.a HC-2000 Formulation

HC-2000 was developed by combining known non-toxic food quality biostimulation accelerants. Ingredients include organic nutrients, proteins, vitamins, co-factors, enzymes, biosurfactants, supplemental carbon substrates, and electron donors. HC-2000 supports native microbial degradation located in the aerobic, facultative, anaerobic, methogenic, and fermentation zones. Raw materials are processed by heated fermentation that is halted prior to completion. This maximizes the concentration of catalysts in a stable, bioavailable, and concentrated form. Two-thirds of the raw materials used in making HC-2000 are distressed fruits that would normally be discarded in a landfill. Converting these materials into a useful product recovers energy and biochemical catalysts that would normally be lost, reduces loadings on landfills, and provides a renewable resource for cleaning up our environment.

During the formulation process, extracellular enzymes and biosurfactants are generated by a proprietary yeast. These two components desorb and convert contaminants into a soluble form for microbial metabolism. In addition, other nutritional components are converted to a form that can be immediately utilized by microorganisms that minimizes cell acclimation times. Acclimation times can take weeks or longer for amendments that are not processed this manner.

HC-2000 provides a multiple catalysts for biostimulation: 1) biosurfactants and enzymes that desorb, break down, mobilize, and prepare contaminants to cross microbial cell membranes for maximum bioavailability; 2) catalysts that drive biochemical reactions in multiple degradation zones; and 3) natural polymers provide slow release mechanisms to minimize amendment wash away.

Sulfates and iron electron acceptors for anaerobic degradation are part of the formulation. Simultaneous addition of multiple electron acceptors (such as Fe^{+2} and $\text{SO}_4^{=}$) increase the total electron accepting capacity to biodegrade many classes of petroleum contaminants. When iron is introduced with other electron acceptors, iron hydroxide clogging precipitates are generally not a problem (1, 6). Nitrates are not part of the formulation (BDL) due to the fact that the drinking water limit is 45 mg/l and are toxic to humans. Sulfites are also excluded (BDL) since they may cause adverse health effects.

Sulfate reduction is the primary pathway for biodegradation of petroleum contamination (6), accounting for 70%, on average, of the natural biodegradation capacity. Sulfate reducing bacteria biodegrade many classes of petroleum hydrocarbons, are highly soluble in water, and tend not to sorb onto soil particles. This increases the storage capacity of electron acceptors that are available to biodegrading microbes. Sulfate amendments are persistent and remain available for use for an extended time. Sulfate microbes are stimulated rapidly (within 45 days after introduction) (18).

The shelf life of HC-2000 concentrate exceeds 10 years if stored at temperatures above freezing and below 120 °F. No nitrates, sulfites, or pathogens are contained in HC-2000.

4.b HC-2000 Regulatory Approvals

HC-2000 has been used on over 400 sites since 1998 including petroleum hydrocarbon remediations in: soil and groundwater, pond sediments, jet fuel and gasoline contaminated wetlands, underground storage tank sites, contaminated stormwater, mineral oil releases in soil and gravel (at power plants and electrical substations), and diesel fuel in railroad ballast. HC-2000 has been approved by the State of Georgia and Florida as an approved site biostimulation agent for soil and groundwater (specific permission required). Federal EPA has granted approval on a case-by-case basis. Several case histories that were approved by regulatory authorities and received *no further action status* are presented herein.

4.c Toxicity Analysis

National Contingency Plan products (NCP) are specifically tested and registered by the EPA for use in saline environments. HC-2000 is designed for use in the more sensitive inland freshwater environment that requires a different testing protocol especially when stimulating native bacteria. EPA testing protocols do not allow for the quantification of the acceleration of native microorganisms present in soil that are the primary degraders. Simply adding amendments to crude oil without native soil microbes provides a potential bias to those NCP list tested amendments containing foreign microbes that can accelerate degradation in a beaker, but do not compete well with the abundant different native species in the various degradation zones in actual field applications. Another complicating issue for NCP testing is that since there is such a wide diversity of native microbial populations it would be difficult to come up with a standard native population that all amendments could be ranked against.

In addition, amendments with 96 hour aquatic toxicities ($\text{LC}_{50\text{S}}$) less than 10 mg/l are banned in California beginning in December of 2016. Several NCP list supplements have toxicities less than, or dangerously close to, this limiting concentration.

To determine appropriate HC-2000 application rates, freshwater aquatic toxicity screens (96 hr. acute static range finding aquatic toxicity tests using an abbreviated ASTM E729-96 with the Trimmed Spearman-Karger Method) were performed. For relative comparison purposes, toxicity tests were run on Microblaze and compared to published toxicity data. Fathead minnows (*Pimephales promelas*) were exposed to five concentrations of Micro-blaze and six concentrations of HC-2000 in freshwater (10).

Tests confirmed that a 6% HC-2000 solution ($LC_{50} = 5,764$) was 4.1 times less toxic than comparable concentrations of Microblaze. In addition, LC_{50} s reported for Microblaze were 1,230 and 1,390 mg/l (20) and compared favorably to 1,414 mg/l during this testing. Aquatic toxicities (LC_{50} s) of select listed products, HC-2000 is 23 times less toxic than the dispersant (Corexit 9500A) used on the recent Gulf oil spill (Deepwater Horizon oil exploration platform on April 20, 2010), 590 times less toxic than BioSolve, and 578 times less toxic than F-500 (Fuel Buster) (Figure 13, Table 2). Note that when a 3% solution of HC-2000 is applied to groundwater, the aquatic toxicity will be considerably less toxic than at 6%.

When applying HC-2000 in soil and groundwater applications at the recommended doses, the food quality ingredients should not adversely effect aquatic life. HC-2000's LC_{50} of 5,784 mg/l aquatic toxicity exceeds 5,000 mg/l is considered relatively non-toxic.

Figure 13: LC_{50} Aquatic Toxicities of HC-2000 Select NCP List Products

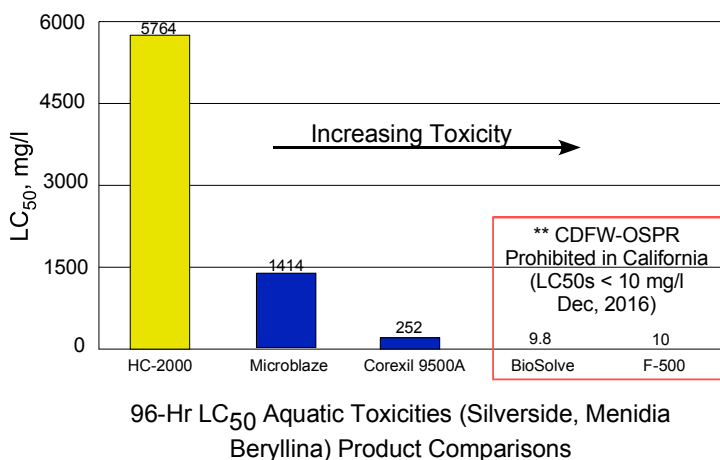


Table 2: Aquatic Toxicity Comparison of LC_{50} s of Select NCP List Products and HC-2000

Agent	LC_{50}
Dispersit SPC 1000	2.9
Nokomis 3-F4	19
Nokomis 3-AA	19
Zi-400	21
Saf-Ron Gold	44
Sea Brat #4	55
Corexit 9500A	130
JD-2000	>5,600
HC-2000	5,764

HC-2000 (HC2) is preserved in a concentrated form at a pH of 3.1 by natural fruit acidity and citric acid (both weak organic acids). Sample HC2 titration curves with top soil (with a high organic content) at a 3% HC2 solution and 6% HC2 solution are presented in Figures 14 and 15 respectively. Top soil with an initial pH of 4.62, 2.2 volumes of 3% HC-2000 can be added while temporary lowering the soil pH to 4.0 (Figure 14). The actual titration curve will vary based on the existing soil, surface water, groundwater, or soil pH and hardness.

Titration 50 mls of sandy clay soil with an initial pH of 6.87 and a 6% solution of HC-2000 required 600 mls to drop the pH to 4.21 (Figure 15). For this pH to be reached, 13.2 volumes of HC2 at 6% were required for one volume of soil. Since HC2 contains weak organic acids, the pH of soils or leachates from soils is not expected to be impacted by more than 1 unit. This is within the optimal pH change (1 to 2 pH units) recommended by EPA for bioremediation (6). Rainfall, stream, or groundwater mixing zones and the hardness of these matrixes should further reduce impacts. Moderate pH adjustment can be made by adding soda ash, if required.

The concentration of sulfate in HC-2000 concentrate is 4,290 mg/l. A 3% solution contains 128.7 mg/l. SO_4^{2-} . This is within the optimal concentration range for sulfate as an electron acceptor (100 to 250 mg/l) to stimulate anaerobic decomposition of the BTEX mass by EPA (3). The federal drinking water standard for sulfate is 250 mg/l based on aesthetic effects (i.e., taste and odor). Georgia does not have stream standards for sulfate or dissolved solids. Water quality standards for sulfate (500 mg/l) and total dissolved solids (1,000 mg/l) have existed in Illinois regulations since 1972. (19).

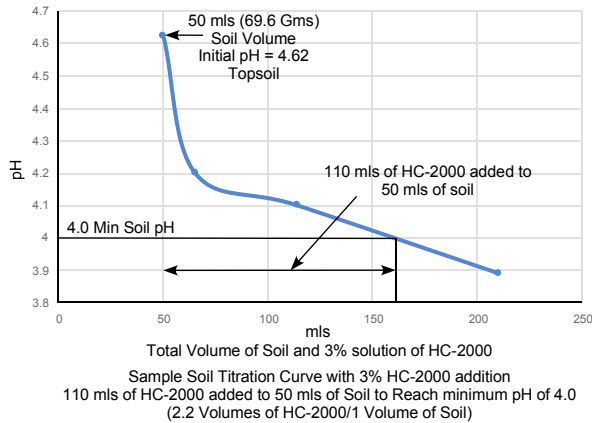


Figure 14: Sample Soil Titration with 3% HC-2000

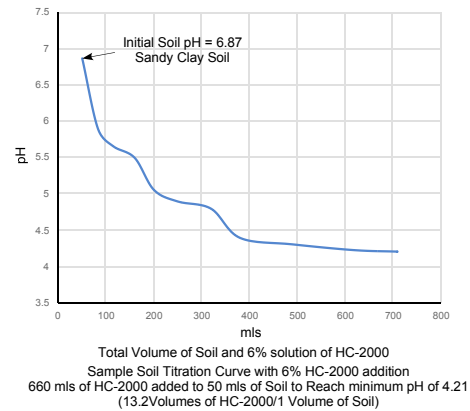


Figure 15: Sample Soil Titration with 6% HC-2000

A 3% solution of HC-2000 contains 1 mg/l Fe²⁺. This is within the optimal concentration range for iron as an electron acceptor (0 to 1 mg/l) to stimulate anaerobic decomposition of the BTEX mass in the anaerobic zone by EPA (6). The current EPA National Recommended Freshwater iron chronic aquatic life concentration is 1.0 mg/l. Considering mixing zones in groundwater systems and iron as an electron acceptor for anaerobic degradation of organic contaminants, iron concentrations should not pose an adverse impact on the aquatic environment.

4.d Efficacy Testing

Biostimulation efficacy testing of HC-2000 was conducted to provide guidance on the effectiveness of HC-2000 in soil and freshwater environments compared to select National Contingency Plan (NCP) products. Parallel efficacy and toxicity tests were also conducted on select products including Micro-blaze, Biosolve, and F-500 (Fuel Buster) for comparison purposes. Tests confirmed that HC-2000 was a more efficient diesel fuel degrader than tested competing products. (Figure 16).

A list of HC-2000 degradable materials listed in references and Remtech full-scale field and bench-scale verified degradable chemicals are presented below (Table 3):

TABLE 3: HC-2000 DEGRADABLE CHEMICALS

LITERATURE REFERENCED DEGRADABLE CHEMICALS

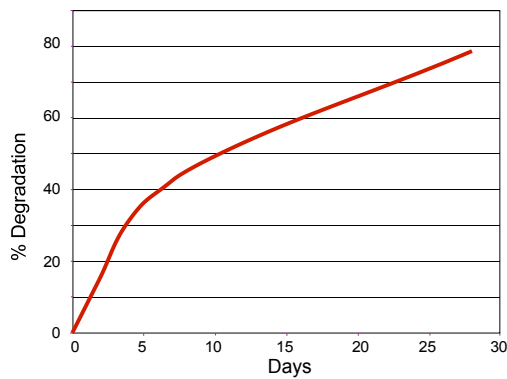
- ▶ Petroleum hydrocarbons
- ▶ Diesel
- ▶ Crude Oil
- ▶ Polycyclic Aromatic Compounds
- ▶ Naphthalene
- ▶ Phenols
- ▶ Halogenated Phenolic Compounds
- ▶ cis-1,2-dichloroethylene (cis-1,2-DCE)
- ▶ 2,4,6-trinitrotoluene (TNT)
- ▶ PCE
- ▶ Polychlorinated Biphenyls
- ▶ Biosorption of Heavy Metals (Cd, Zn, Pb, Cu, As, Hg, Cr, U, Th, Mn, Fe, Ni, Co)

REMTECH VERIFIED DEGRADABLE CHEMICALS

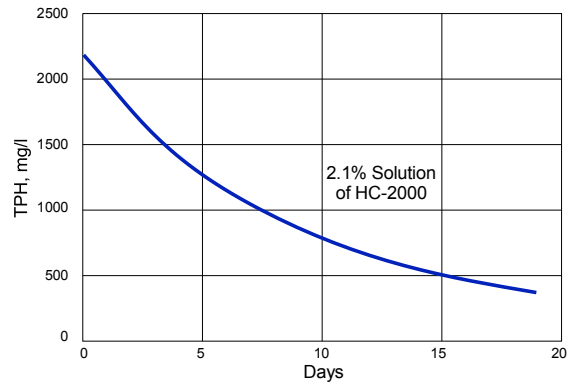
- ▶ Petroleum Hydrocarbons^b
- ▶ Diesel^b, Jet Fuel^b, Jet A^b, JP-4^b
- ▶ Gasoline^b
- ▶ Polycyclic Aromatic Compounds^b
- ▶ PAHs^b
- ▶ Benzene^b, Creosote^a
- ▶ Toluene^b, Ethylbenzene^b, Xylenes^b
- ▶ Crude Oil^a
- ▶ Motor Oil^b
- ▶ Mineral Oil/Transformer Oil^b
- ▶ TCE^a, PCE^a, PCBs^a
- ▶ Ethylene Glycol^a

^a pilot test verified

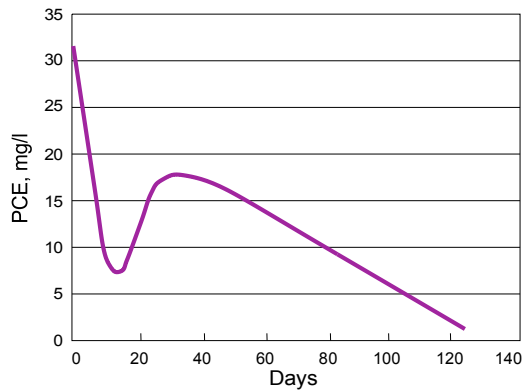
^b full-scale project verified



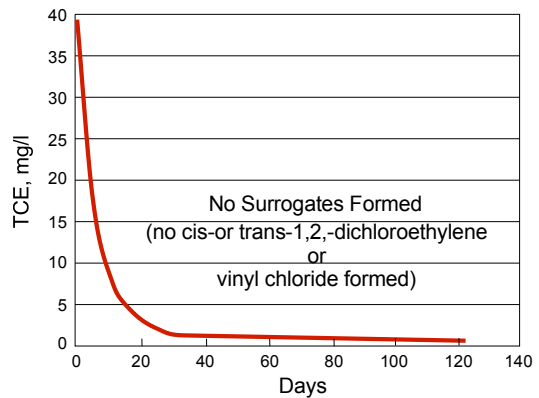
78% TPH (Crude Oil) Removal in 28 Days Aerobic Slurry Reactor



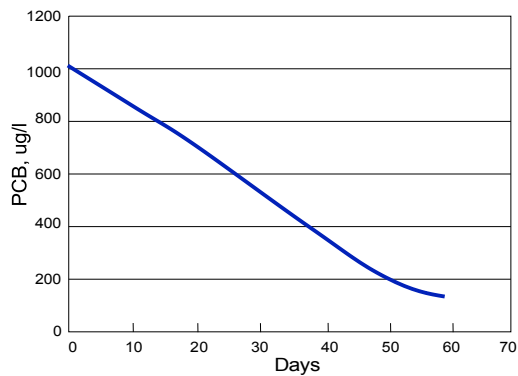
86.8% TPH (Motor Oil & Creosote) Removal in 19 Days Aerobic Slurry Reactor



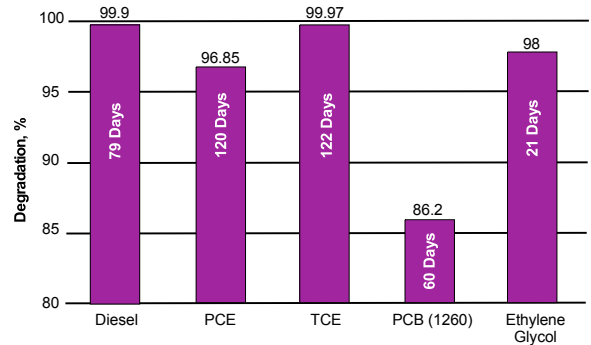
96.85% PCE Removal in 120 Days Aerobic Slurry Reactor



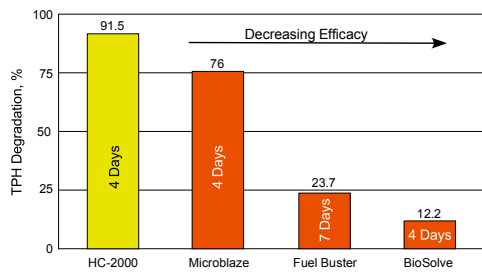
99.97% TCE Removal in 122 Days Aerobic Slurry Reactor



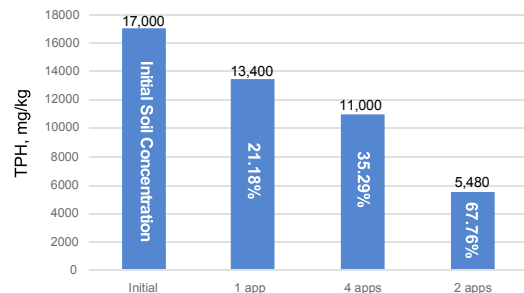
86.2% PCB Removal in 60 Days Aerobic Slurry Reactor



Bench-Scale Soil Slurry Analyte Degradation/Time Comparisons



Four (4) Day Bench-Scale Soil Slurry Diesel (TPH) Degradation Product Comparisons (Fuel Buster - 7 Days)



HC-2000 Mineral Oil/Transformer Oil Soil Degradation Over 9 Week Period Number of HC-2000 applications (app) over Treatment Period

Figure 16: HC-2000 Bench-Scale Select Chemical Efficacy Tests (10)

4.1 HC-2000 SOIL & GROUNDWATER BIOREMEDIATION APPLICATIONS

4.1.1 Soil and Groundwater Workflow Diagram

A sample workflow diagram for HC-2000 soil and groundwater bioremediation applications is presented in Figure 17.

The objective of **site characterization** is to determine the feasibility of bioremediation. Investigations should begin with review of site history, activities associated with property occupants, raw materials used, waste products produced, and disposal practices at the time. Historical records of wastewater disposal frequently indicate how and where waste was discarded. Review of historical soil or groundwater investigations or remedial actions, according to current understanding and experience, on similar bioremediations can be useful to provide information on the evaluation and selection of potential bioremediation strategies.

For instance, military bases have been a dumping ground for used solvents to clean parts, fuels, and corrosion inhibitors. Used solvents and fuel were burnt in test pits that were used for fire training.

Some petroleum companies sold filling stations to third parties to divest themselves of potential liabilities associated with leaking underground storage tanks. Dry cleaners spilled perchloroethylene, lead smelters used lead and acids and frequently buried waste, refineries had waste ponds with solvents and heavy metals, and power plants stored ash with heavy metals and coal with contaminated leachates. Landfills have the potential for hazardous waste leachate migration and vapor intrusion.

Looking at historical aerials and interviews with past employees can identify potential areas to investigate. Sandborne maps can identify locations of USTs.

In one the author's experience while being mentored by corporate directors of environmental affairs for several major chemical companies including Monsanto, Dow, Diamond Shamrock, and others, it was required that *an industry's operations and raw materials needed to be understood to better predict what contamination is likely to be present in order to come up with applicable remediation strategies.*

Smart Investigations should be conducted where test borings and wells can be converted or used as part of the treatment or monitoring systems. Direct interpretation by professional remediation experts in the field while borings and test data are being gathered can minimize the amount of data required to predict the best potential remedial strategy that will work at a specific site.

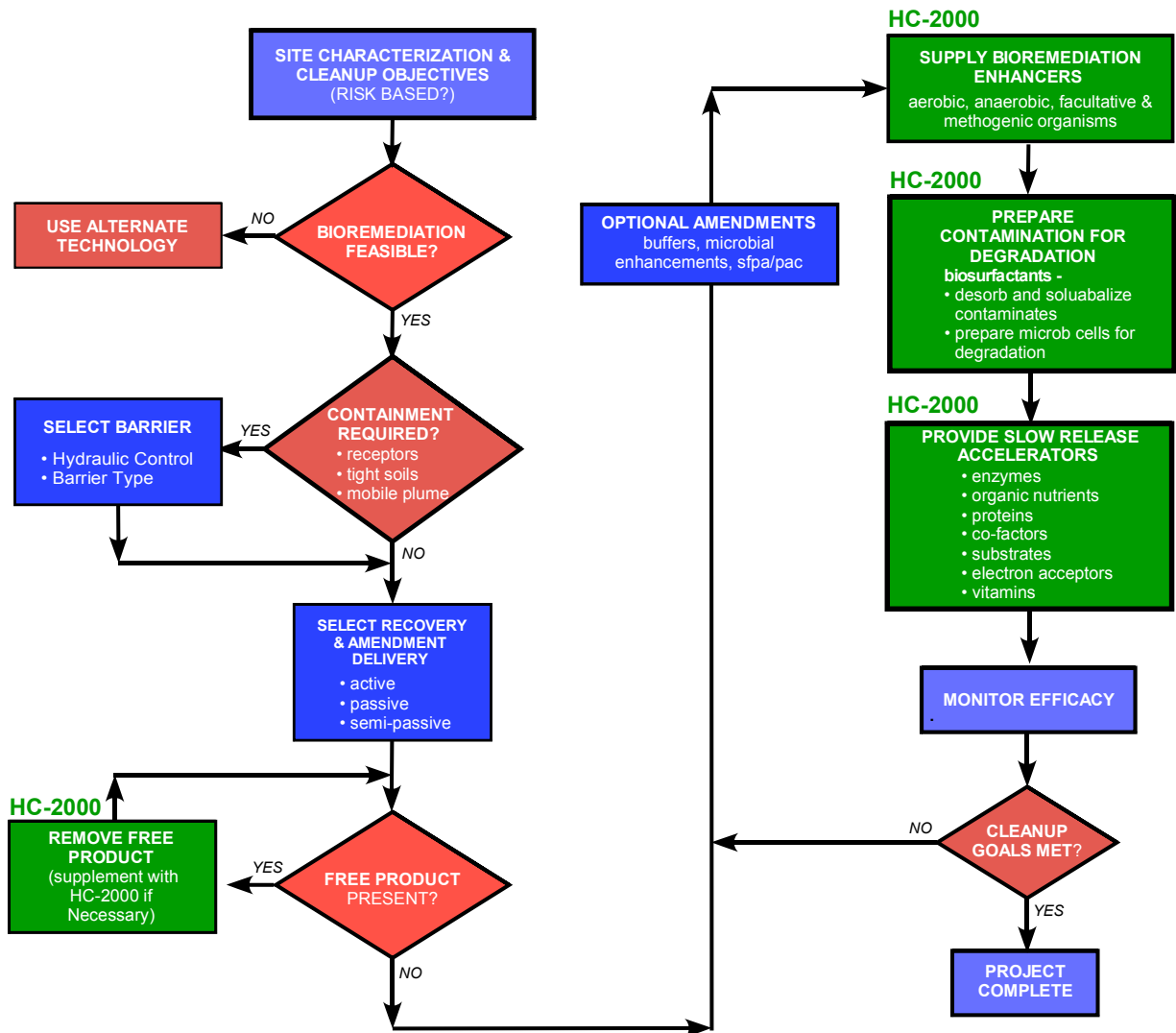
Experience on past remediations can identify potential strategies early in the investigation. Direct observations by experienced professionals can predict qualitative estimates that can be later fine-tuned and quantified. Site characterization data should include conductivity, soil classification, depth to groundwater, flownet, LNAPL, DNAPL, type and degradability of contamination, presence of free product, solubility, sorbability, location of contamination, microbial plate counts, and pH. Treatment system grid design should be determined by radius of influence determinations via vacuum, air pressure, tracer testing, or other testing techniques.

Determine Cleanup Objectives - The next step is to rank the risk of the site based on potential imminent health hazards, site location relative to sensitive receptors, contaminant(s) location, toxicity, mobility, mass, and soil classification, conductivity, in order to identify **cleanup objectives**. Can they be risk based?

Consider Other Remedial Solutions - Bioremediation can potentially be ruled out if any of the following exist:

- Are imminent health hazards present?
- Will unachievable cleanup standards be required?
- Are shallow groundwater tables less than 10 ft present?
- Do conductivities less than 10^{-4} cm/sec exist?

Figure 17: HC-2000 Bioremediation Soil & Groundwater Bioremediation Workflow Diagram



Consider Other Remedial Solutions, cont.

- Are low plate counts present?
- Is the groundwater pH outside the range of 5 to 9?
- Are excessive concentrations of LNAPL or DNAPL present?
- Are other remedial alternatives available for conductivities between 10^{-4} and 10^{-6} cm/sec?
- Is free product or contamination overload present for microbial degradation?
- Can LNAPL or DNAPL be removed efficiently and cost-effectively based on the site characterization?
- Are soil petroleum hydrocarbon concentrations exceeding 50,000 ppm or solvent concentrations exceeding 7,000 ppm present?
- Will soil washing with biosurfactants or generation of biosurfactants following amendment addition reduce mobile phases to manageable residual levels while minimizing the potential for rebound of back-diffusion?

Barriers or Hydraulic Capture Required? Leachability and transport of contamination, amendments, location of contamination source, and location relative to sensitive receptors influences the decision whether or not to consider if barriers or hydraulic control are required.

Document Baseline Conditions. Collect baseline geochemical, biochemical, and contaminate concentrations and integrate with the bioremediation treatment program.

Select and Install Delivery and Monitoring System. This should be based on site characterization. Install passive, intermittent, or active system. Use hydraulic controls, barriers, or amendment recirculation, if required.

Monitor Efficacy of Degradation. Use monitoring wells to monitor effectiveness of degradation, amendment concentrations, formation of secondary degradation products, and potential mobilization of metals or the generation of hydrogen sulfide or other secondary degradation byproducts.

Adjust Treatment to Field Conditions. Adjust or modify amendment delivery method, concentrations, add buffers, or supplemental amendments as needed. Note that HC-2000 has natural polymers present that facilitates the introduction of hydrophobic SFPAC carbon into solution for transport through the aquatic environment. Continue process until cleanup goals are met.

Bioremediation May be Complete if cleanup goals are met or if asymptotic contaminant concentrations have been achieved allowing treatment to be suspended. Risk based closure may be considered if the contaminant plume has been immobilized or contained. Additional monitoring may be required to ensure rebound or back-diffusion does not occur or is low enough to allow natural attenuation to degrade contaminants to acceptable levels.

Suggested Dosage Rates

Optimal degradation conditions are present when total heterotrophic plate counts or specific degrader counts are elevated and maintained during the treatment period. Elevated plate counts indicate that sufficient nutrients, moisture, and environmental conditions are present.

Secondary parameters that may be monitored include: respiration by-products, moisture, dissolved oxygen, pH, total dissolved solids, TOC, and redox potential. Monitoring the reduction of the contaminant(s)-of-concern determines when treatment is complete and/or when natural attenuation can complete the degradation process.

The *rules of thumb* dosage for HC-2000 range from one gallon of HC2 concentrate to three (3) to ten (10) cubic yards of contaminated media (soil, groundwater, railroad ballast, etc.) over the treatment period. Treatment periods range from five weeks to several months. Concentrate dilution ratios of one part of HC-2000 to sixteen parts of water (6% solution) is recommended for soil, ballast or gravel applications. Groundwater applications are recommended at an application rate of 3%. The number of applications and concentrations may vary according to site specific requirements. Care should be exercised to prevent over dosing with HC2 to prevent native microbes from utilizing HC2 rather than the targeted contaminant.

Groundwater dosage rate of 1:30 (3%) is a typical starting point that should be confirmed. Always add HC-2000 concentrate to water to avoid excessive foaming.

Typical injection rates in groundwater are at least 0.25 to 0.5 gallons per minutes at pressures less than 10 psi. Several applications of multiple pore volumes are generally required to achieve cleanup objectives.

Case History 1: Bioremediation of JP-8 Spill in Wetlands

A tanker carrying 8,000-gallons of JP-8 on a major interstate highway rolled down a forested embankment releasing 2,600-gallons of jet fuel into an area immediately adjacent to a stream buffer bordering a golf course in North Atlanta.

Excavation was ruled out due to potential stream bank and highway destabilization due to sharp slopes and high-density forestation holding the bank in place. Steep creek banks were severely eroded due to highly fluctuating stormwater flowrates from encroaching rapid urban development and overloaded stream capacities. Insitu enhanced bioremediation was selected over other remedial strategies due to limited site access and significant cost savings over other methods, i.e. less than 50% of site excavation and restoration costs.

This setting was ideal for insitu bioremediation due to highly permeable soils and favorable biological degradation mechanisms including rhizodegradation, phytodegradation, phytovolatilization, phytostabilization, transpiration, air stripping, and redox biodegradation reactive zones.

A cutoff membrane wall was installed at the plume's leading edge. 700-gallons of free product were recovered during the first month from fourteen (14) interceptor trenches with horizontal screens and vertical risers.

HC-2000 injections were initiated by surface spraying, injections through 10 biosparge points, and recirculating amendments through 6 interceptor trenches. Leachates with amendments were recovered for recirculation from downgradient horizontal screens. The leachate treatment train consisted of an oil/water separator followed by granular activated carbon filtration.

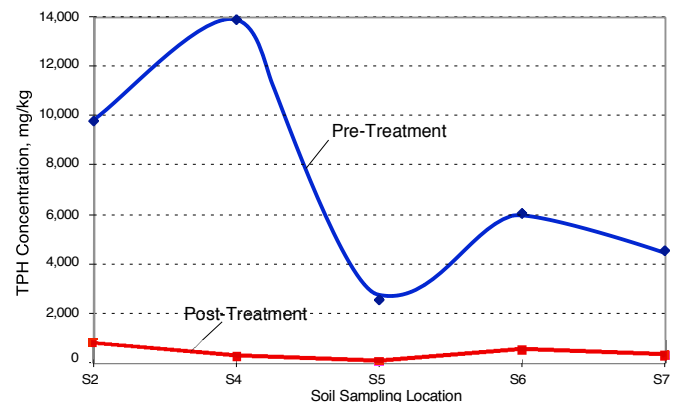
Groundwater dissolved oxygen levels were elevated from 0.5 mg/l to over 3.5 mg/l. Total groundwater heterotrophic plate counts increased from 10,000 CFUs/ml to over 1,000,000 CFUs/ml. Intermittent treatment continued for a four-month period.

Soil TPH concentrations were reduced over 91% and groundwater TPH concentrations by over 94%. Benzene groundwater concentrations were reduced from 125 ppb to below detection limits. Groundwater benzene concentrations remained below detection limits six (6) years after the incident.

Downstream concerns, including the golf course, attempted to file a multimillion dollar law suit against the insurance company based on erosion damage. Baseline documentation of area conditions prior to initiation of remedial activities and the fact the remediation was done insitu with minimal disturbance served as a basis for suite dismissal. Existing erosion was due to rapid urban development with increased runoff that overloaded area streams.

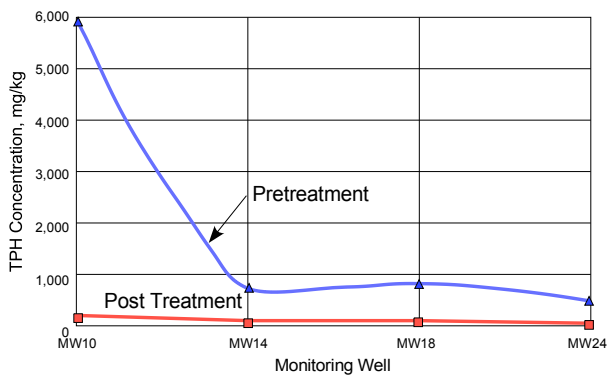
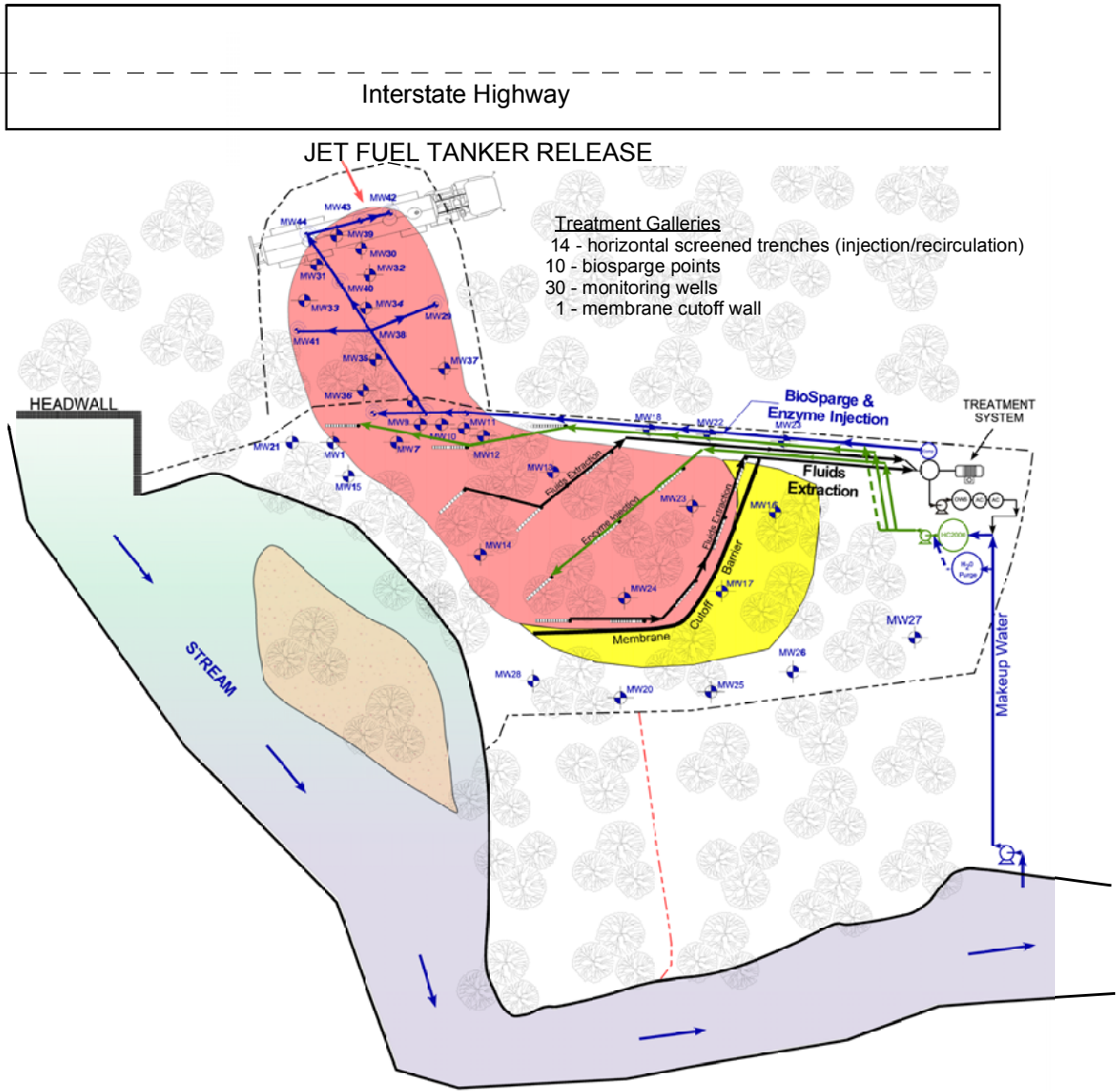


HC-2000 Treatment Galleries

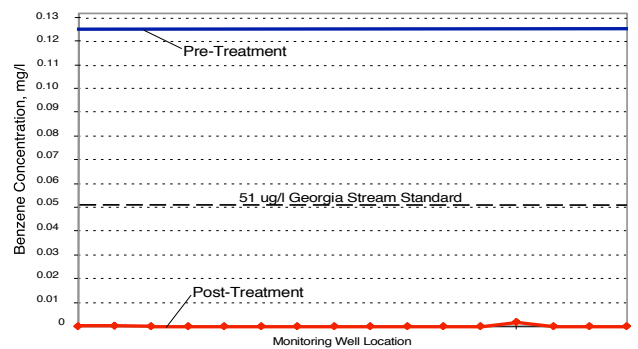


Soil TPH Concentrations reduced from 91 to over 99% Following HC-2000 BioVenting

Jet Fuel Bioremediation Treatment Grid



JP-4 Tanker Spill Wetland Groundwater Recirculation Bioremediation in Four (4) Months



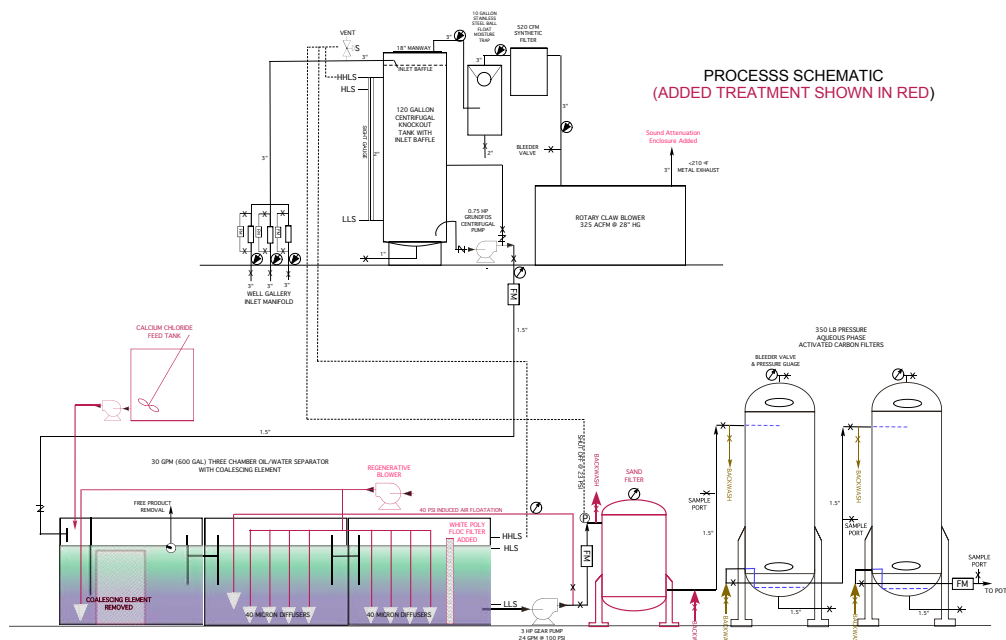
Groundwater Benzene Concentrations Reduced Below Detection Limits at Monitoring Wells Following BioSparging

Variable densities of this biomass resulted in solids remaining in a stabilized suspension resisting separation. The heaviest biomass was experienced closest to wash bay 1 with declining concentrations 50 ft downgradient. Fuel separation from the biomass in the oil/water separator (OWS) was achieved by incorporating dissolved air flotation and calcium chloride addition (coagulant and flocculant aid). Sand filter media was replaced with anthracite and pea gravel to facilitate more efficient filter backwashing prior to carbon filtration. Entrained fuel biomass removed from the OWS were transferred to three 500-gallon holding tanks that provided additional fuel/solids separation after a 30-day retention period.

The system was operated in an intermittent pulsed fashion by running extraction wells on timers and applying vacuum to six recovery wells at a time in an attempt to minimize biofilm formation. The state initially required removal of free product prior to amendment addition.



Remtech Manufactured Dual-Phase Extraction Treatment Trailer



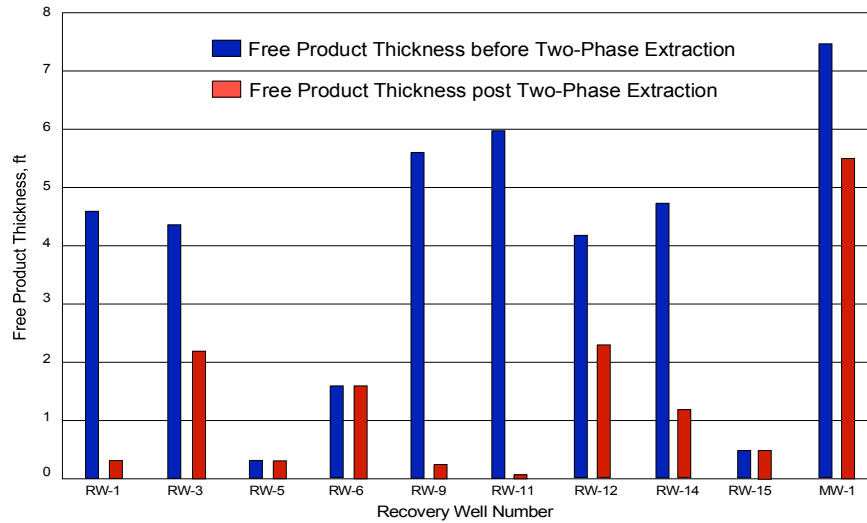
Four years into the project, an estimated 48.44% of the free product had been recovered. Free product thickness persisted in wells ranging from 0.34 ft to 7.5 ft. Permission was finally granted to inject HC-2000 with its biosurfactants and bioremediation catalysts to attempt enhance the removal of the free product phase.

Several well rehabilitations were conducted during the project to attempt to kill and reduce the amount of biofilm generated. The rehab included surge blocks, jetting, brushing, hydrochloric acid, calcium hypochlorite, hydrogen peroxide, citric acid, glycolic acid, and HC-2000. HC-2000 proved to be the most effective. Unfortunately, the sludge production rate was consistent throughout the duration of the project.

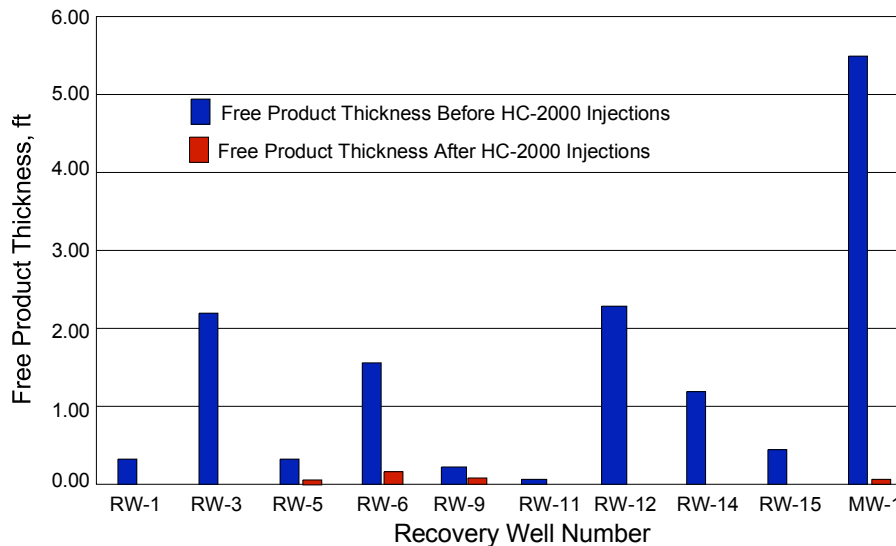
Fuel entrained biomass was chemically fixed with Portland Cement. Chemical fixation released additional free product that was combined with other recovered fuel that was sent to a recycler. Biomass production rates were some of the highest experienced in the state.

Seven HC-2000 injections over a one-year period resulted in a 92.63% reduction of the remaining free product. Each injection was followed by a water chase to transport amendments into the formation and away from wells.

A “no-further action letter” was issued, based on a state site risk assessment, concluding that the mobile phase had been effectively removed and with sensitive receptors located several miles downstream.



48.44% Free Product Thickness Reduction in Extraction Wells Following Four (4) years of Dual-Phase Extraction.



92.68% Free Product Thickness Reduction Following One (1) Year of Dual Phase Extraction with HC-2000 Injections



Heavy Iron Bacteria in OWS From Wells Closest to Bay 1

Case History 3: Gasoline LUST BioSparge Site

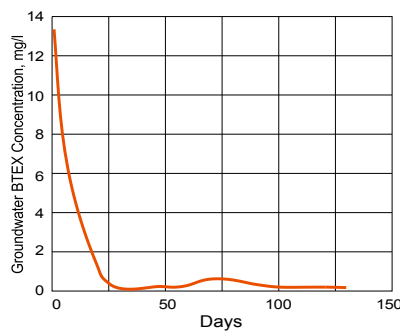
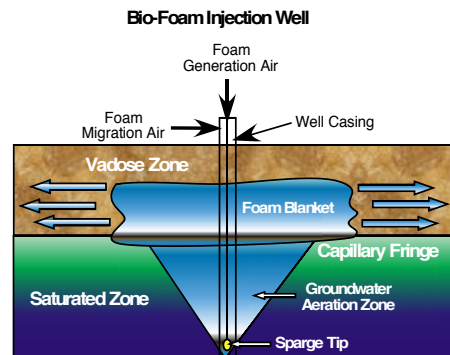
After four years of bioventing and groundwater pump and treat at a Georgia LUST FUND site, seven existing recovery wells at a beverage distribution facility site were converted into HC-2000 biosparge injection wells. The hydraulic conductivity at this site was 2.2×10^{-4} cm/sec.

HC-2000 was injected over a six-month period. Biosurfactants contained in HC-2000 formed a foam blanket that increased the mass transfer of amendments through the saturated zone. BTEX concentrations in the heart of the groundwater plume were reduced by over 94% and TPH concentrations were reduced by over 96% during the treatment period. Free product was eliminated during the first month of treatment.

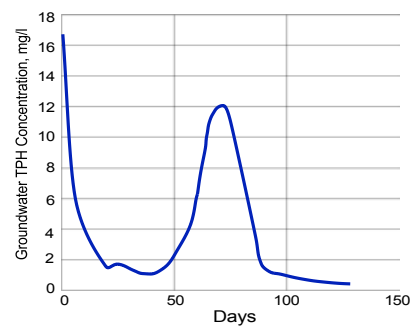
LUST GASOLINE SITE BEVERAGE DISTRIBUTION BIOSPARGE REMEDIATION (NO FURTHER ACTION)



BioFoam Injection Well Head



BTEX Reduced by 94% After 120 Days in Heart of Spill



TPH Reduced by 96% After 120 Days in Heart of Spill

Case History 4: UPS UST Gasoline Tank Pit Groundwater BioSparge Bioremediation

A gasoline underground storage tank pit was contaminated with dissolved gasoline analytes following removal and backfill of a 10,000-gal UST. This residual contamination prevented site closure.

Sixteen geoprobe biosparge points were driven into a gravelly clay soil. Remtech's native bioremediation accelerator HC-2000 was injected topically and through the sparge tips on a weekly basis. An automated compact rotary vane air compressor and HC-2000 (HC2) injection system was used to pulse air and a 3% solution of HC2 into the tank pit.

At the end of a six-week treatment period, total heterotrophic plate counts jumped to several hundred million CFU/ml. Groundwater BTEX analytes were reduced - 98% for benzene, 96% for ethylbenzene, 99% for toluene, and 94% for total xylenes.

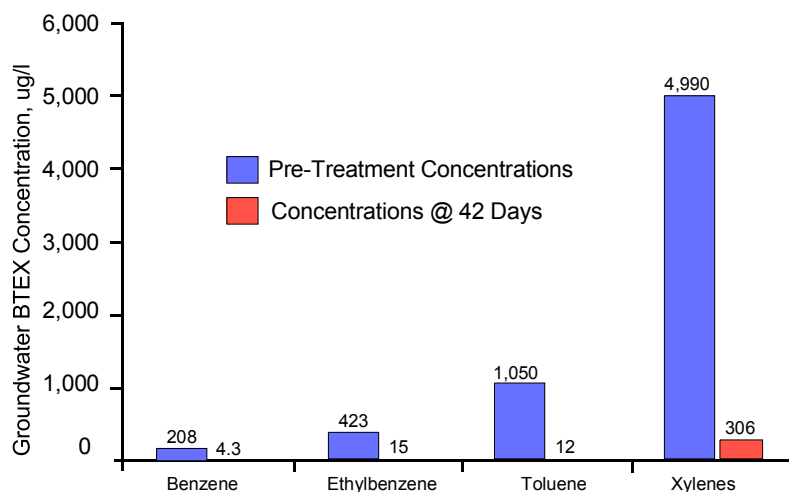
The tank pit was closed, minimizing additional facility operational interruptions.



16 Biosparge Points in UST Tank Pit



Remtech Fabricated Biosparge Rotary Vane Air Injection System



BTEX Reduced by 96.8% during Six (6) Week Treatment Program

Case History 5: International Airport 1,000 Gallon Jet A Fuel Spill Bioremediation

The drive train on a tanker carrying 10,000-gallons of Jet A exploded and punctured the shell releasing 8,000 gallons of fuel on a perimeter road and into a drainage ditch. The site was located in a restricted area in direct line with aircraft approach runways.

Fuel was released on asphalt pavement and migrated overland and one mile downstream into a drainage ditch, through tunnels, and a creek. The airport authority required that a remedial approach with minimal access and visual disturbances to approaching aircraft be implemented.

Approximately 6,500 gallons of fuel were recovered from a one-mile section of stream utilizing skimmers, vacuum trucks, wash down pumps, and a polypropylene rope mop.

An estimated 1,000 gallons of fuel saturated 600 cubic yards of soil at the release point. 350 cubic yards of soil were excavated from the bottom of the drainage ditch and placed on a one foot deep biobed. Soil was aerated with a disc/harrow.

The remaining 250 cubic yards of soil were treated insitu employing three horizontal biovents and a regenerative blower. Ten gallons of HC-2000 (diluted with 16 volumes of water) were applied on a weekly basis to the bank and biobed. A sprinkler system was installed to maintain soil moisture levels at both locations.

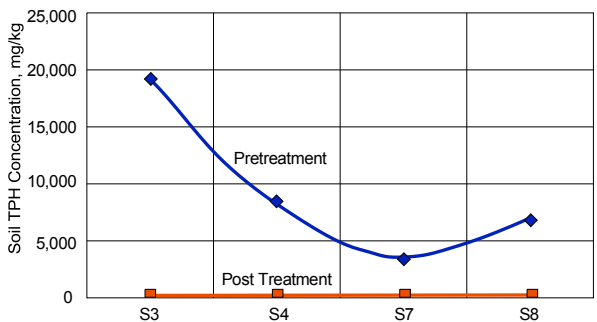
Total petroleum hydrocarbon (TPH) concentrations were reduced below 200 mg/kg in 16 weeks (99% reduction). Total heterotrophic plate counts were elevated from 6,000,000 to over 100,000,000 CFU/gm during the treatment program.



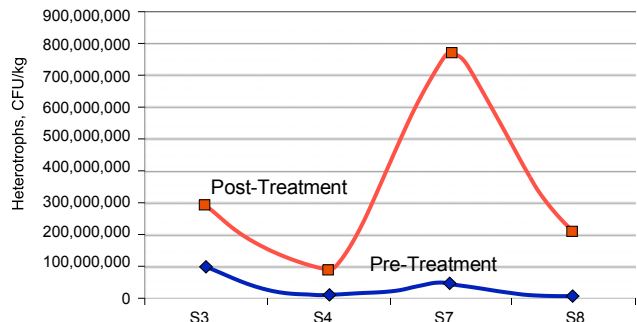
Soil Biobed



BioVenting System with HC-2000 Injection



TPH Concentrations Reduced by Over 95% after 4 Months



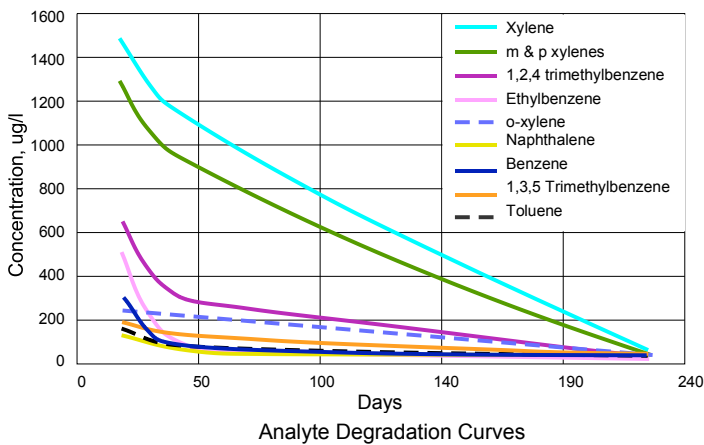
Total Heterotrophs Increase with HC-2000 Applications

Case History 6: HC-2000 Bioremediation of Solvent Site, Albany, NY

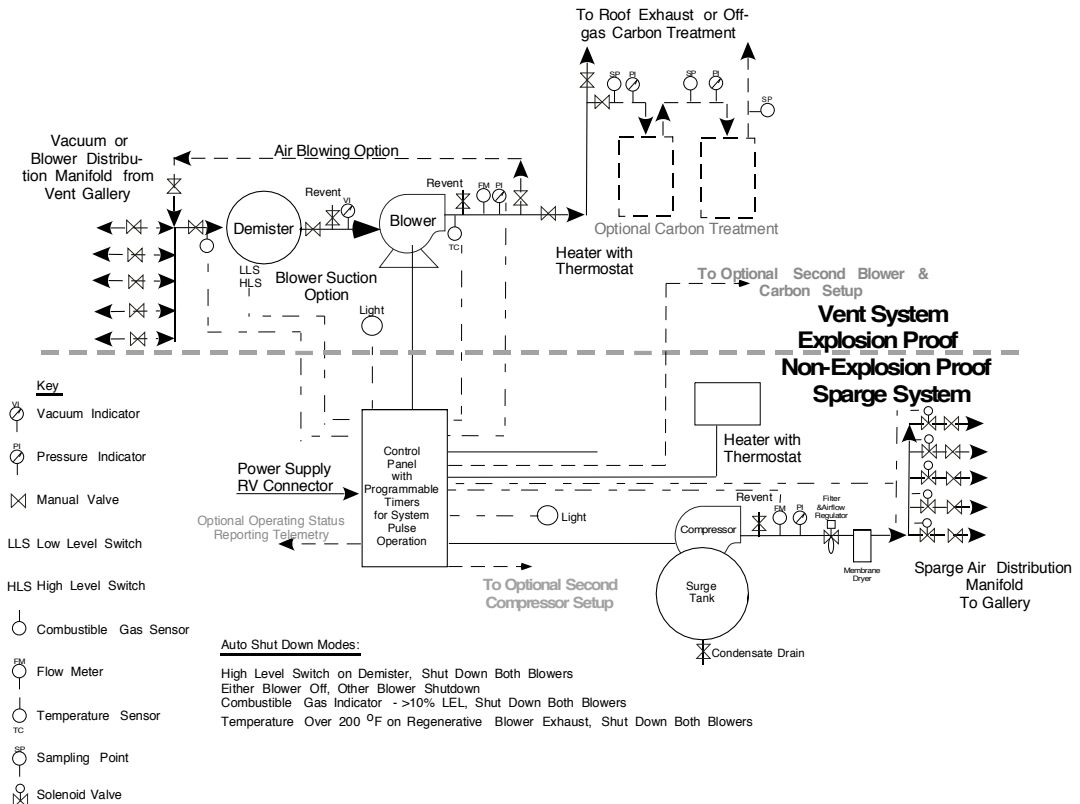
Soil and groundwater under an 8,000 square foot beverage bottling plant and parking lot was contaminated with non-chlorinated solvents. Remtech was engaged to manufacture a mobile treatment system to deliver HC-2000 at multiple facility sites.

Remtech manufactured a UL certified mobile bioremediation trailer. This system was equipped with telemetry reporting. Remtech installed five air sparge wells and five horizontal vapor extraction wells to enhance treatment under the building and parking lot.

The site geology consisted of sandy clay and glacial till with a shallow groundwater table (3 feet BLS). After 5.5 months of system operation, analyte concentrations were reduced: benzene - 100%, ethylbenzene - 100%, naphthalene - 95%, toluene - 95%, 1,2,4-trimethylbenzene - 97%, 1,3,5-Trimethylbenzene 85%, and total xylenes - 98%.



Remtech Manufactured Biosparge & Bioventing Trailer

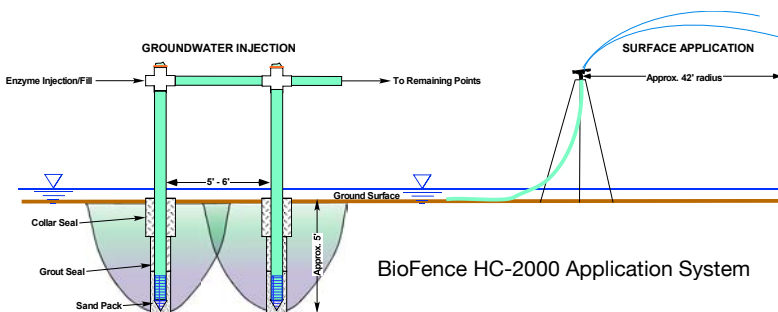
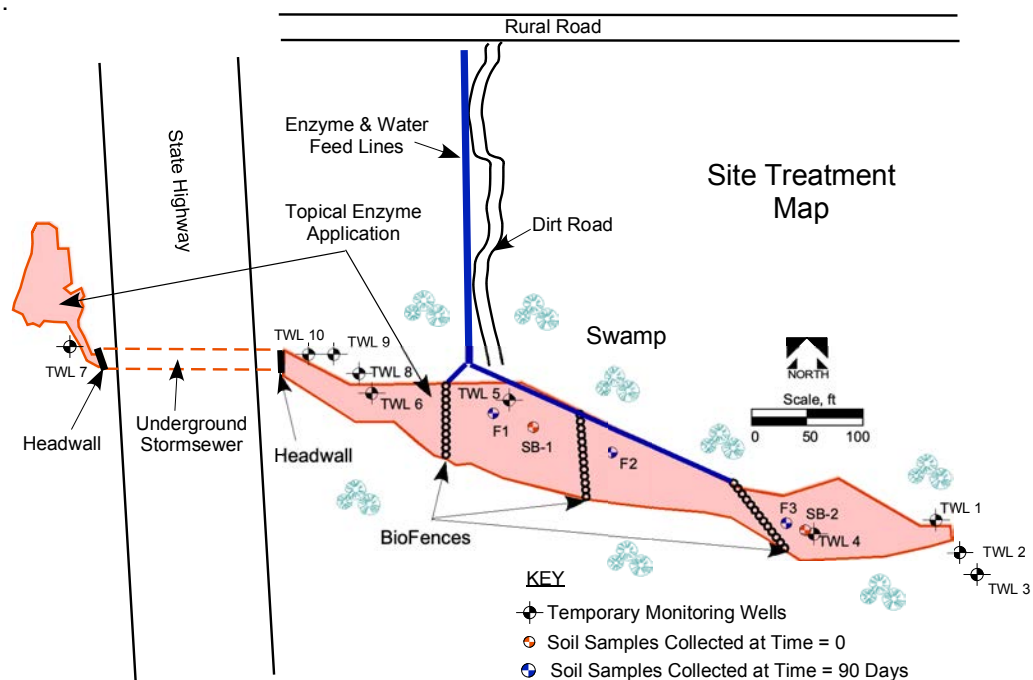


Case History 7: Flooded Lowland Gasoline Spill Bioremediation

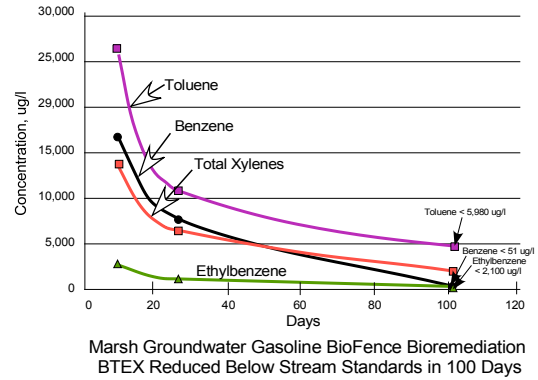
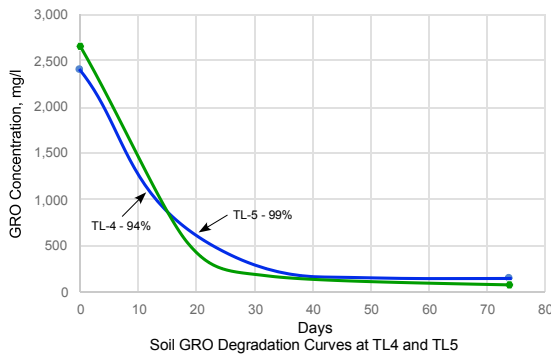
A gasoline tanker rolled over in a seasonally flooded lowland releasing an estimated 2,500 gallons. The gasoline plume migrated over a 0.56 area on a pine tree farmer's property. Excavation was ruled out due to inaccessibility, area flooding, and soft soils. The Georgia Department of Natural Resources had the transportation company contact Remtech Engineers to employ its proprietary native bioremediation accelerator HC-2000.

Three biofence reactors consisting of 30 injection points and a sprinkling system were installed allowing topical and subsurface HC-2000 application. HC-2000 was applied at one to two weeks intervals followed by a one-hour watering period. HC-2000 migrated through the contaminated media via advection and diffusion between injection points. Soils in this vegetated area were classified as loamy sand to a depth of three ft, followed by sandy clay loam at depths to 6 ft BLS. Soil and groundwater samples were collected and analyzed for TPH (GRO) and BTEX respectively. Rhizodegradation, phytodegradation, and phytovolatilization were contributing bioremediation mechanisms. 386 gallons of HC-2000 were applied to 3,613 cy of soil or 1.1 gal/10 cy of soil plus a volume of water that passed through the flooded area.

This project was completed in twenty weeks. Total heterotrophic bacteria plate counts increased from 290,000 CFU/gm to over 5,000,000 CFU/gm during the treatment process. Average soil Gasoline Recoverable Organic concentrations were reduced 96.5% and average groundwater benzene concentrations were reduced by 99.89%. Remtech received a *no further action letter* from the Georgia Department of Natural Resources upon project completion. No damage to site vegetation was observed.



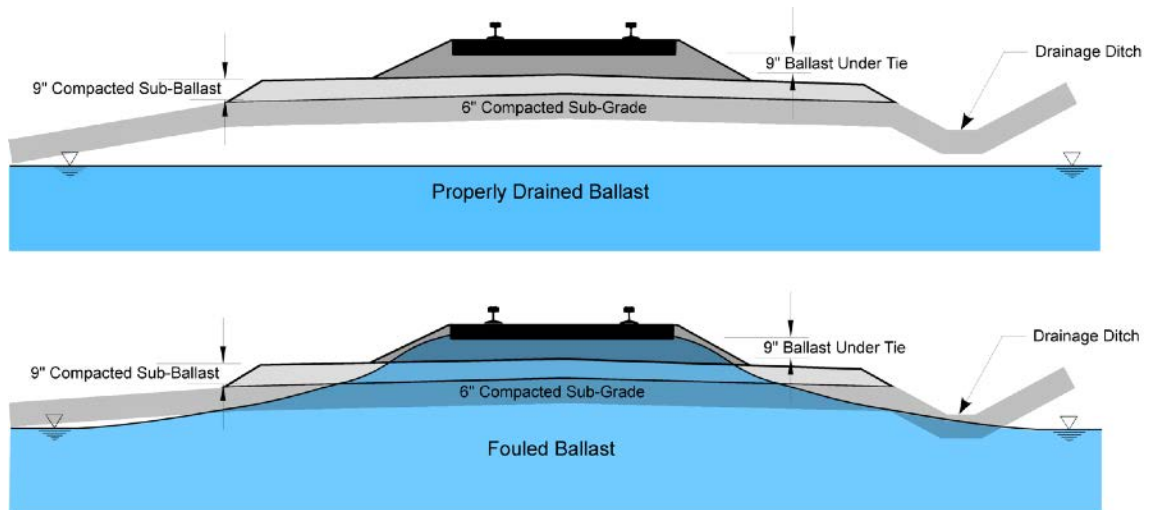
BioFence Injection System



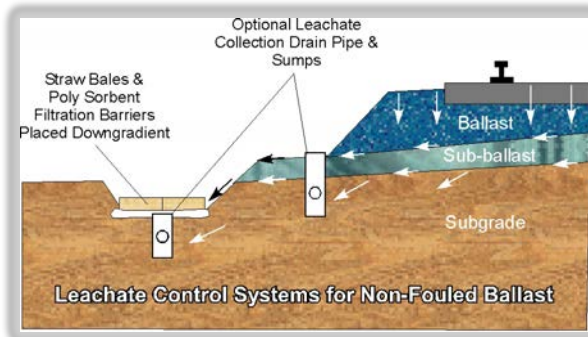
4.2 HC-2000 RAILROAD BALLAST APPLICATIONS

HC-2000 has been used effectively on diesel fuel and lubricant spills on railroad tracks from derailments, sideswipes, engine and track equipment fires, and at-grade collisions with minimal interruptions of track traffic. A typical workflow diagram is presented in Figure 18.

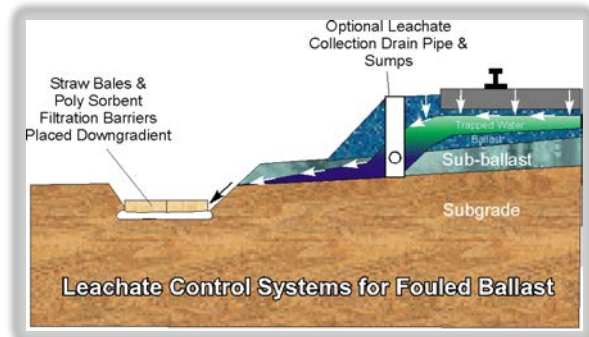
Track beds with large ballast covering compacted fines serve as a biological trickling filter. Railroad ballast contains 59 to 94% of all organisms that are petroleum hydrocarbon degraders. Bacteria have become acclimated due to deposits of lubricant oils comprised of long-chain hydrocarbons including: machine grease, fuel oils, and transformer oils that contribute to the presence of polycyclic aromatic hydrocarbons (PAHs) and TPH. Background TPH concentrations of 1,000 mg/kg have been observed in ballast. HC-2000 is also an effective degrader of PAHs and other railroad related oils.



Two types of track beds are generally present: fouled ballast and non-fouled ballast. Fouling is caused by breakage of ballast aggregate, spillage of coal dust from moving trains, decaying weeds, and migration (or pumping) of subgrade particles into surface materials. As the percentage of fouling increases, more water is retained, resulting in the buildup of pore water pressure. Ballast is designed to provide free drainage with particle sizes ranging from 0.3 to 2.5 inches. Fouling reduces void spaces and restricts drainage.



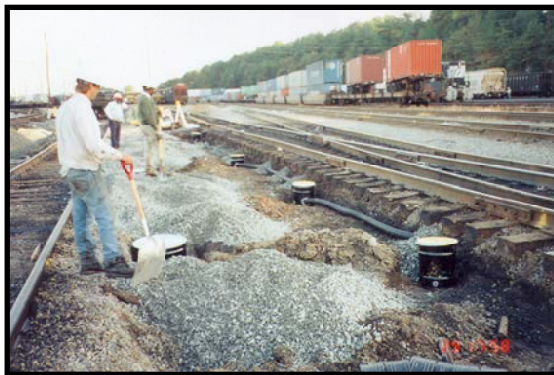
Non-Fouled Track Leachate Collection System



Fouled Track Leachate Collection System

Trapped water in fouled track beds limits the vertical migration of NAPL. Hydrophobic petroleum hydrocarbons either pool or migrate away from the track bed according to the local topography. Test pitting and trenching can determine the location of free product and assist in installing a product recovery system.

Installation of interceptor trenches with drainage tile, sumps, and straw bales with polypropylene sorbent boom placed in drainage ditches can provide collection systems for intercepting free product and controlling leachates during the treatment period. A combination of HC-2000 followed by a water chase can accelerate the desorption of petroleum hydrocarbons from the track bed. Additional applications of HC-2000 can be made following removal of the bulk free product. During the degradation phase, total heterotrophic plate counts typically increase from millions to billions of CFU/gm.



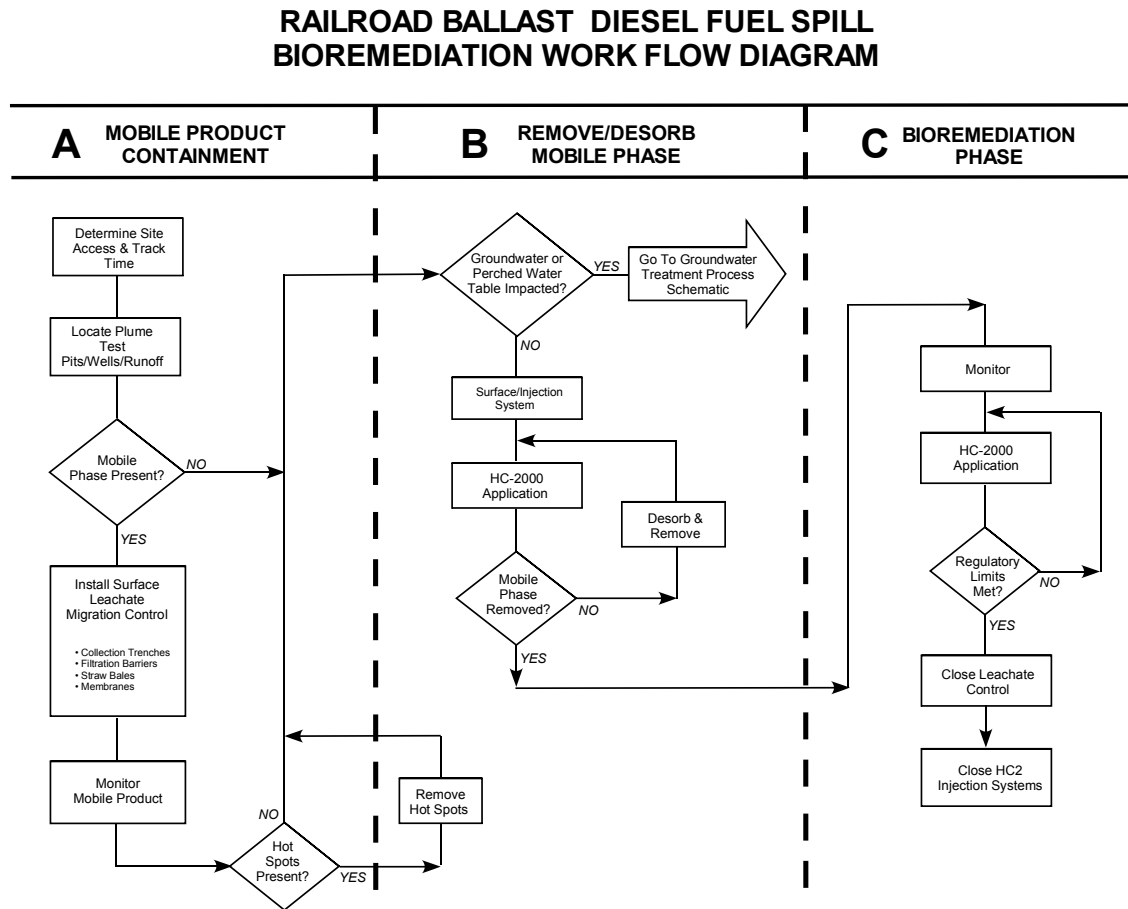
Leachate Perforated Pipe and Sump Collection System



Desorption of Lube Oil from Ballast with HC-2000

In non-fouled track beds, petroleum hydrocarbons can migrate through the ballast, sub-ballast and into the subgrade. If enough product is released, groundwater can be impacted. The initial remedial approach can be the same as for a fouled track bed with followup test borings/monitoring wells to establish the location of the plume. Soil and groundwater bioremediation strategies can be implemented if groundwater is impacted (Figure 17).

Figure 18: HC-2000 Track Ballast Petroleum Hydrocarbon Bioremediation Workflow Diagram



4.2.1 Railroad Ballast Workflow Diagram

A. Phase 1: Locate, Contain, and Monitor Mobile Phase

- Determine site access and available site service interruption time, i.e., track time, underground utilities, environmental receptors, sensitive environments and structures, and cleanup and risk-based targets.
- Locate contamination and mobile phase pathways. Look for short circuits, i.e., sewers, swales, drains, backfill, utilities, and streams using test pits, monitoring wells, and geoprobe points.
- If mobile phase is present, install containment or free product migration monitoring system. Install leachate/migration control systems (straw bale filtration dams, collection trenches, pits, drainage tiles and sumps, or temporary detention pools). For track applications, install leachate control near toe of ballast and/or in drainage swales. Apply HC-2000 followed by oxygen saturated water chase to increase penetration, moisture, and oxygen.
- Protect structural integrity of track, grounding, and cathodic systems - Do not disturb ballast below ties or within a 45% slope of tie edges.
- Monitor and contain free product movement.

B. Phase 2: Remove/Desorb Mobile Phase

- Remove mobile phase or saturated soil/solids hot spots and define HC-2000 injection method and grid.
- If perched or groundwater is impacted go to the soil and groundwater work flow diagram (Figure 17).
- Apply HC-2000 (followed by water chase to accelerate removal of mobile phase and assist with product desorption). Bound contaminants may account for over 60% of contamination. Fuels may become mobile as they are desorbed and broken down into shorter hydrocarbon chains.
- Ballast Treatment – Apply HC-2000 topically followed by a water chase or through injection points. For fouled ballast - mixing or agitation enhances HC-2000 delivery and treatment. Pressurized water (3,000 to 5,000 psi) or a cribbing bucket to loosen ballast next to ties may be used.
- Repeat HC-2000 applications until mobile phase dissipated.

C. Phase 3: Bioremediation Phase

- Monitor leachates (visual observation in stormwater runoff - no sheens), sample ballast fines and downgradient soil (at edge of railroad R.O.W.) and leachate for total petroleum hydrocarbons. Supplemental water may be required to maintain moisture levels at 70% of field holding capacity.
- Apply HC-2000 per label instructions and adjust for site conditions.
- Repeat until regulatory limits, risk-based closure, or until natural attenuation can reach regulatory limits.
- Remove/close leachate control systems.
- Close HC-2000 injection systems.

Case History 8: Track Siding Lube Oil Bioremediation

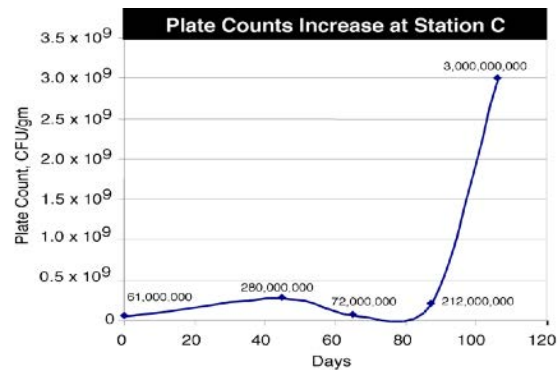
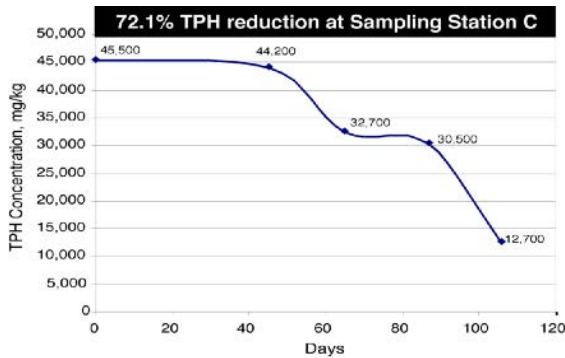
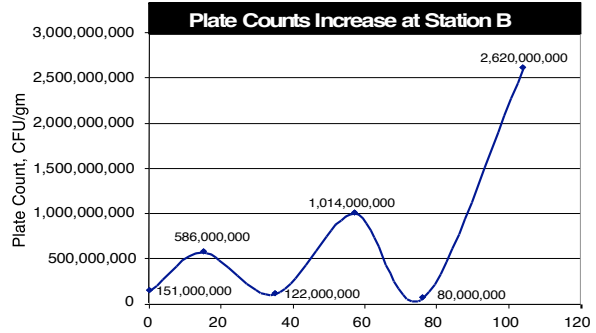
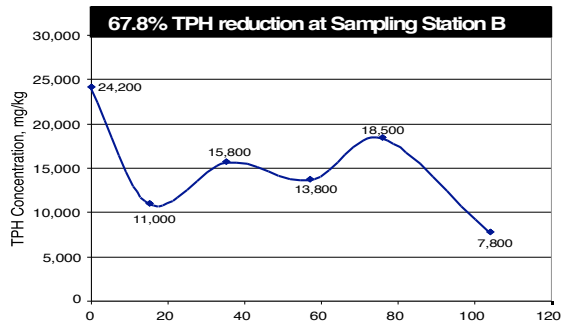
A lubricant oil distribution facility accumulated lube oil on its track siding over a period of 30 years. Releases were from railcar transfer leaks to above ground storage tanks.

The contaminated track ballast was 164 ft long x 15 ft wide by 3 ft deep. Ballast was tilled with a cribbing bucket between the ties. HC-2000 was applied topically over a four (4) month period. Total petroleum hydrocarbons were reduced by over 60%. Further reductions would have been achieved with a longer treatment program. Lube oils are one of the more difficult petroleum hydrocarbons to degrade. HC-2000 biosurfactants and enzymes broke down the long-chained hydrocarbons into shorter chained molecules that were easier to degrade.

The client began using track mats and spill pans to reduce further transfer releases. The rail car shipper was also required to stop overfilling railcars and clean cars off prior to shipping to cut down on releases during offloading.



30 Years of Lube Oil Accumulation on Ballast Reduced by Over 60% After Four (4) Months of HC-2000 Treatment



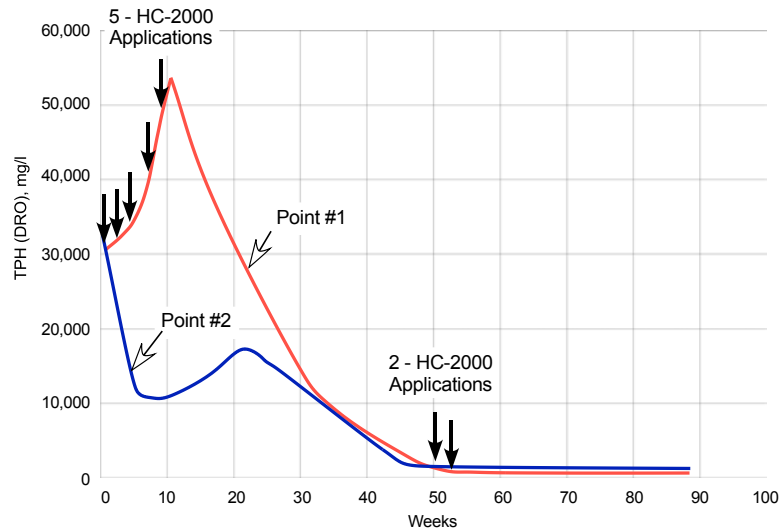
Case History 9: Railroad Ballast Diesel Spill Fuel Bioremediation

A 1,000-gallon diesel fuel release from an at-grade collision with a truck was broadcast over 1,800 ft of a track siding. Seven surface applications of HC-2000 were applied to the railroad ballast over a period of 52 weeks.

Biosurfactants in HC-2000 desorbed the fuel from the ballast to track bed fines where native heterotrophic bacteria were present. Biochemical catalysts reduced DRO TPH concentrations in the track bed by 97% at Impact Point 1 and 99% at Resting Point 2. The track was monitored for an additional 35 weeks and no TPH rebound occurred. A *no further action letter* was obtained from the State Department of Natural Resources.



Application of HC-2000 to Track Siding



TPH Concentrations Reduced by an average of 98% over a period 50 weeks with HC-2000

Case History 10: 1,000 Gallon Diesel Spill Bioremediation of Track Siding

An at-grade crossing collision on a track siding with a train and fuel tanker released over 1,000 gallons of diesel fuel over a track distance of 175 feet.

The fuel saturated asphalt road and sub-grade were removed and replaced. An initial application of HC-2000 and water chase were used to flush the mobile fuel phase out of the ballast. A total of twelve applications of HC-2000 (205 gallons) were made over a twelve-month period. Degradation monitoring continued for fourteen months. A drinking water well was located less than 500 ft downgradient from the release. A monitoring well was installed immediately downgradient from the tracks to ensure that the water well was not impacted.

No contaminants were detected in the water well. Groundwater Polycyclic Aromatic Hydrocarbons (PAHs), BTEX, and (DROs) were degraded below detection limits in the monitoring well. Half of the costs associated with this project were due to demonstrating that this release did not impact the well.

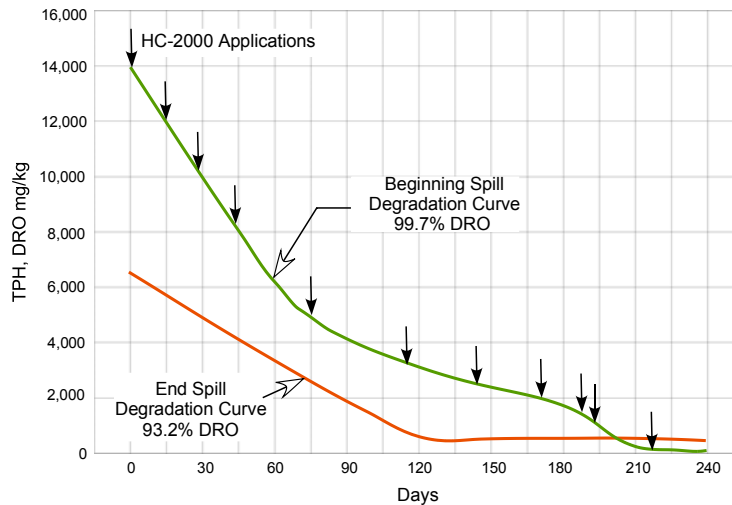
Diesel Range Organics (DRO) ballast/soil concentrations were reduced by an average of 96.5%. A *no further action letter* was received from the State.



HC-2000 Application to Diesel Impacted Ballast



Site Following Bioremediation Completion



Railroad Ballast DRO Degradation Curves

Case History 11: Railroad Track Waste Oil Cleanup

A waste oil recycler tanker spilled an estimated 600 gallons of waste oil that flowed through a storm drain and onto two main line tracks covering 140 ft of drainage ditch and 60 ft of track.

Six HC-2000 applications were made during a four-month period. Leachate collection and filtration system maintenance and monitoring continued for an additional eight-month period until sheens in stormwater runoff and oil collection in the leachate recovery system dissipated. Concentrations of residual waste oil were reduced to levels for natural attenuation to continue.



Leachate Collection System Installed Between Tracks



HC-2000 Application to Waste Oil on Ballast



Oil Leachate Collection Sump

Case History 12: Bioremediation of Locomotive Fire on Mainline Track

A locomotive caught fire on a major railroad's super elevated mainline track. An estimated 100 gallons of diesel fuel were released along a 100-foot track section.

Remtech designed and installed a solar/battery powered water mist system to supply oxygen and water to the ballast during the remediation process to maintain a 75% water field holding capacity.

Track ballast served as a biological trickling filter. HC-2000 was applied during the first three days of treatment followed by weekly HC-2000 applications to accelerate the native fungi and bacteria biodegradation process. Air sparge tips were driven into the ballast to increase oxygen levels. A battery driven compressor was utilized to supply air through the sparge tips.

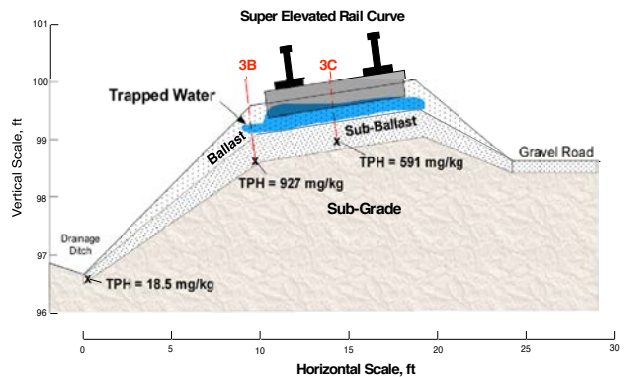
Four sampling points were monitored over a 118-day treatment period. Average TPH concentrations were reduced 90%. TPH concentrations at the toe of ballast next to the drainage ditch were recorded at 18.5 mg/kg at the end of the project indicating that petroleum hydrocarbons were being degraded within the railroad ROW.



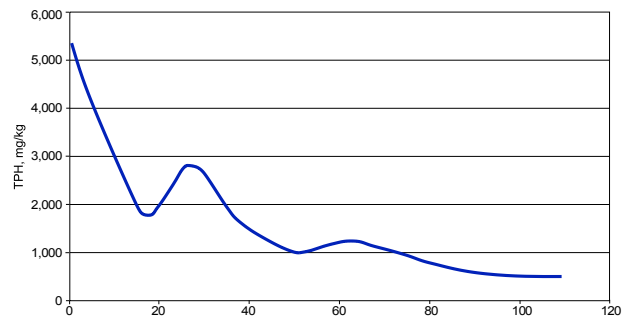
Solar Powered Water Mist Application



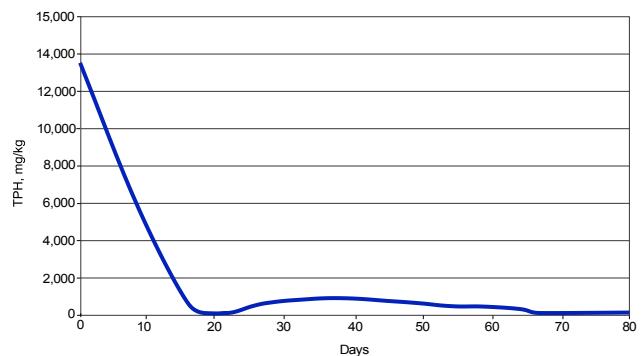
HC-2000 Track Bed Application



Cross Section Through Sample Location 3



Sample Point 3C



Sample Point 4B

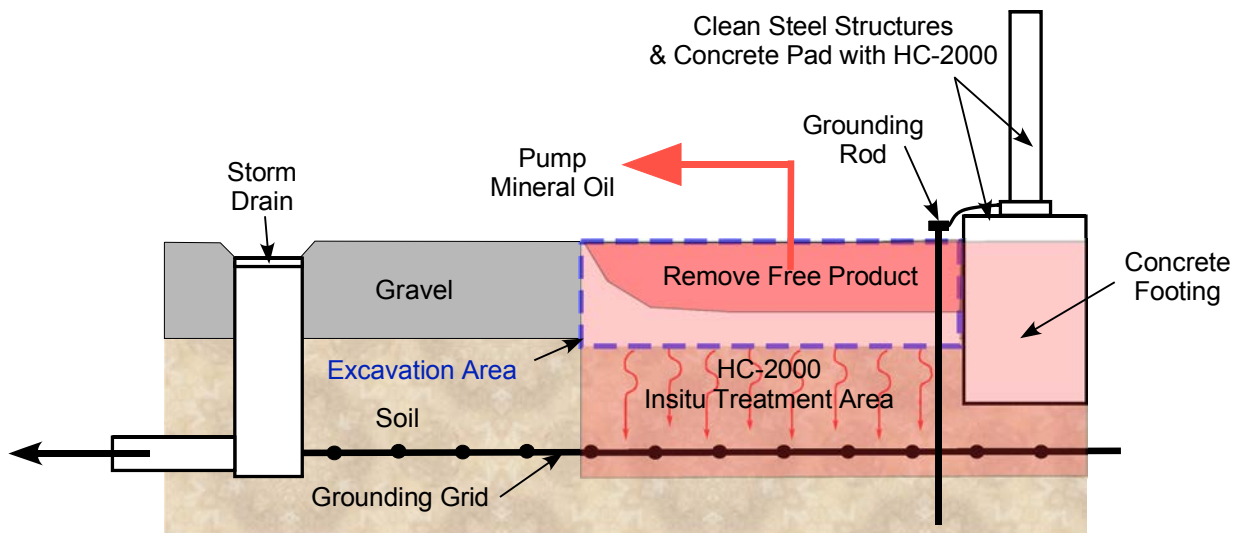
4.3 POWER PLANT AND SUBSTATION MINERAL OIL RELEASE BIOREMEDIATION APPLICATIONS

Mineral oil releases can occur from substation fires originating from oil-insulated circuit breakers, reactors, regulators, current transformers, power transformers, DC valves, oil-insulated cable, hot work procedures, arson, smoking, lightning, storms with downed power lines, metal piercing bullets from hunters, and other mechanical impacts. Internal arcing can produce breakdown gases that can build up pressure causing insulating oil to breach enclosures resulting in a release.

PCB insulating oils have been largely eliminated since they were banned in 1977. The Toxic Substances Control Act regulates PCBs. Transformers that contain PCB's are required to be labeled and releases should be remediated according to TCSA and EPA requirements.

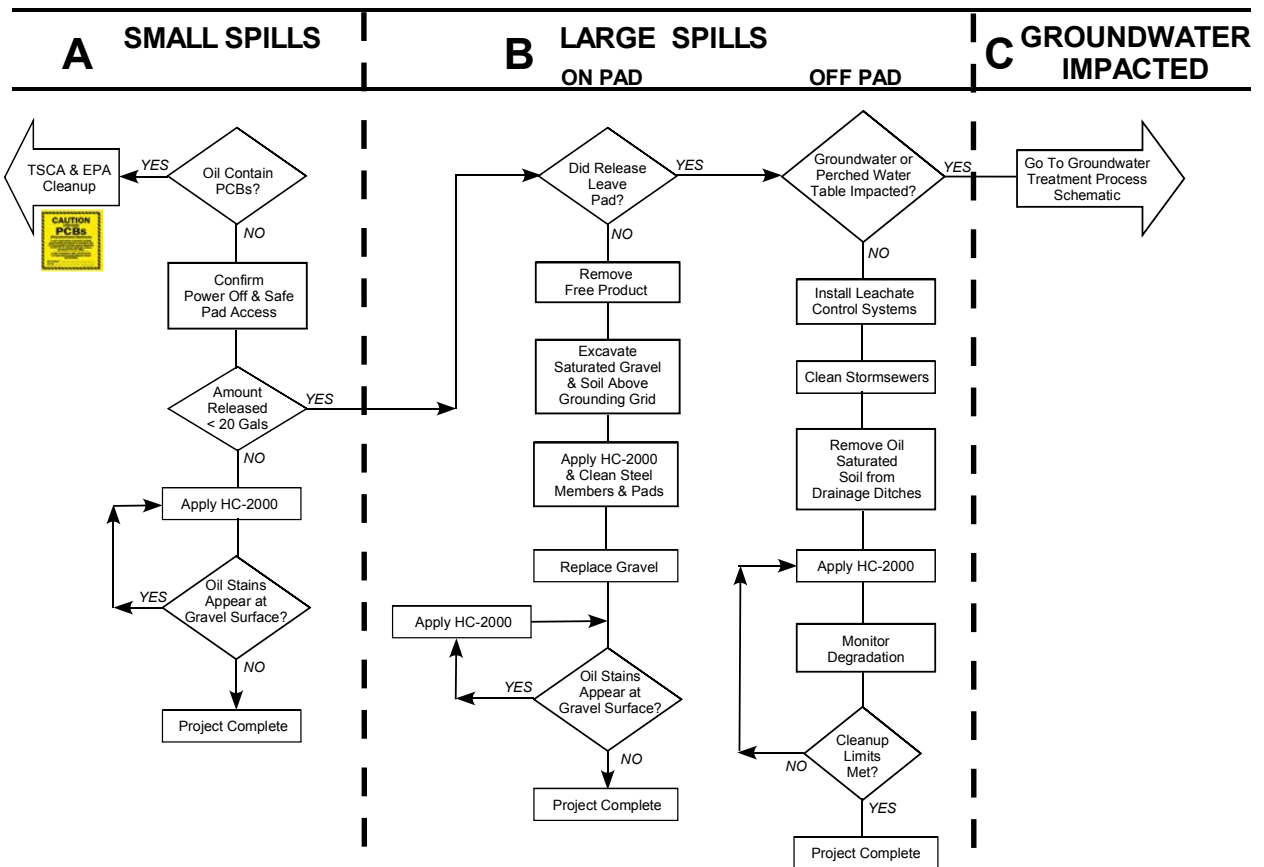
Substation pads are designed to keep water away from electrical equipment. Pads are usually graded flat with gradual slopes towards fence-lines. Soil bases under gravel are generally compacted clay that minimizes oil penetration. The majority of oil spills are contained in the surface gravel.

HC-2000 is designed to treat PCB free small mineral oil contaminated gravel/soil and larger releases in the grounding grid zone after removing grossly contaminated soil and gravel. HC-2000 can also be used to clean oily metal surfaces and concrete footings/pads for electrical equipment. Oil sheens are stopped during the treatment period. A sample work flow diagram is presented in Figure 19.



Substation Pad Excavation and Grounding Grid HC-2000 Treatment Zone

Figure 19: HC-2000 Substation Mineral Oil Bioremediation Workflow Diagram



4.3.1 Mineral Oil Release Treatment Workflow Diagram

HC-2000 is designed to treat PCB free mineral oil small spills and larger releases in the grounding grid zone after removing grossly contaminated soil and gravel. HC-2000 can also be used to clean oily metal surfaces and concrete footings/pads for electrical equipment. Make sure that PCBs are not contained in mineral oil. Cleanups with PCBs must comply with the Toxic Substances Control Act.

A. Small PCB-Free Mineral Oil Spills

- Small mineral oil spills (<20 gallons) can be sprayed with HC-2000 using a Hudson sprayer. Mineral oil is washed down into fines under gravel with biosurfactants contained in HC-2000 where native heterotrophic bacteria degrade oil to carbon dioxide and water. Power must be turned off prior to applying HC-2000.
- Biosurfactants also break down long-chained mineral oils into smaller molecules that are easier to degrade. Bioremediation catalysts in HC-2000 accelerate the degradation rates and provide nutrients and accelerants that are released slowly to provide longer amendment contact time.
- Sunlight causes mineral oil to form dark stains. Degradation is considered complete when stains on gravel surfaces disappear. In some cases, several applications of HC-2000 may be required to remove staining. Oil sheens are stopped during the treatment period.

B. Large PCB-FREE Mineral Oil Spills

- For spills greater than 20 gallons, oil will generally pool or fill voids in the gravel. Power must be turned off prior to applying HC-2000.
- Pooled oil should be removed by pumping or using a vacuum truck. Oil saturated gravel and soil above the grounding grid is excavated and disposed of.
- Soil above and in the grounding grid zone is treated with HC-2000 to degrade residual contamination. Steel supports and concrete slabs supporting electrical equipment may be cleaned by brushing or pressure washing with HC-2000.
- Storm drains and drainage ditches must be checked to determine if oil has migrated off the substation pad. Free product and oil saturated soil should be excavated and disposed of prior to applying HC-2000.
- Leachate containment barriers such as straw bales or poly sorbent boom in drainage ditches should be installed until the degradation program is complete. HC-2000 is applied topically to soils with a fire or garden hose nozzle that will not clog from pulp.
- Repeat applications of HC-2000 until cleanup requirements are met. Typical total petroleum hydrocarbon concentrations for residential cleanups are less than 100 mg/kg and for industrial sites 1,000 mg/kg. Confirm cleanup targets with local regulatory authorities.

C. Groundwater Impacted

- If mineral oil impacts perched water tables or groundwater, proceed to the groundwater treatment work flow diagram (Figure 17).



Remove Free Product and Saturated Gravel



Excavate Saturated Soil Above Grounding Grid



Clean Metal Superstructure and Treat Grounding Grid Soil with HC-2000



Replace Gravel

Case History 13: Substation Transformer Explosion Bioremediation

A 7,000-gallon transformer exploded at a power substation. Dielectric fluid was released to the gravel/soil pad. Mineral oil also migrated into stormwater drainage ditches. The release was located within 500 feet of a private drinking water well. Remtech was engaged by the power transmission company to remediate the site. Free liquids were pumped into tankers and sent to an oil recycling facility.

Mineral oil saturated gravel and soils located above the grounding grid system were excavated. Residual contaminated soils in and below the grounding grid system and groundwater were treated insitu with Remtech's HC-2000 natural bioremediation accelerator.

Monitoring wells were installed to track plume migration and the accelerated degradation of the mineral oil in soil and groundwater. No contamination was detected in the drinking water well located downgradient of the substation during a 12-month degradation and monitoring period.

Insitu treatment of transformer oil contamination located in and below the substation's grounding grid system with HC-2000 allowed the substation to be put back in service rapidly, minimizing power interruptions, and without removal and replacement of the grounding system.



Transformer Oil Release



Closeup of Transformer Oil Release

Case History 14: Pole Mounted Transformer Oil Bioremediation

Excessive rain caused the root structure of a two-foot diameter tree to fall over, knocking down a power pole with three transformers, causing a power outage to a major commercial complex. The transformers fell in a drainage ditch with three rock check dams leading to a stream 400 ft downgradient.

An estimated 40-gallons of mineral oil were released during a rainstorm in the early morning hours. Rain saturated grass limited the vertical penetration of oil allowing free product recovery with poly propylene sorbent. The grassed drainage ditch with utilities was sprayed with HC-2000 to desorb additional oil. Additional oil was recovered on sorbent booms and



3 Transformers Landed in Drainage Ditch Releasing 40 Gal of Transformer Oil

Over a nine week degradation period, two applications of HC-2000 degraded the mineral oil 67.76%. A parallel pilot test was conducted to illustrate that overdosing with HC-2000 can have a retarding affect on degradation rates, i.e., 4 applications of HC-2000 over the same treatment period only had an efficacy of 35.29% indicating that native microbes were utilizing HC-2000 as a food source rather than targeting the contaminant of concern.



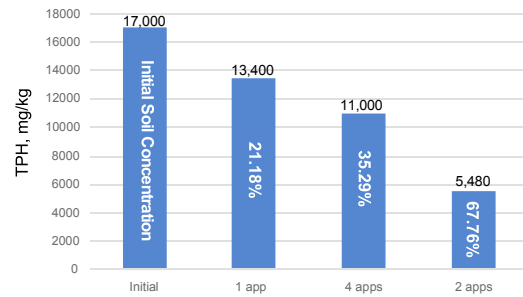
Polypropylene Sorbent Removes Bulk Mineral Oil



HC-2000 Sprayed on Grass to Desorb Additional Oil and Accelerate Degradation



Site Monitored Through Three Additional Rain Events Until Oil Sheen Dissipated in Stormwater Runoff



HC-2000 Mineral Oil/Transformer Oil Soil Degradation Over 9 Week Period
Number of HC-2000 applications (app) over Treatment Period

Case History 15: Substation Regulator Explosion Mineral Oil Bioremediation

A PCB-free 385-gallon regulator at a substation was punctured by a bullet that resulted in an explosion with an ensuing fire on New Year's Day with the entire contents released. One hundred gallons of pooled mineral oil was recovered with diaphragm pumps from the saturated gravel. Mineral oil also migrated beyond the substation's fence line through a 160 ft storm drain that flowed into a drainage ditch that joined a creek 1.2 miles downstream. Two hundred and twenty-five feet of drainage ditch were impacted by this release.

Saturated soil and gravel above the grounding grid was excavated and loaded into a 20 cy rolloff box for disposal at a Subtitle D Landfill as a special waste. Oil-saturated soils, immediately below the stormdrain outfall, were excavated and combined with soils in the rolloff.

The stormdrain was cleaned and purged with a 6% HC-2000 solution followed by several water chases. Rinsates were collected and combined with the mineral oil for a total of 200 gallons of recovered liquids.

Brush was removed from the 225 ft long ditch and polypropylene sorbent was used to remove free product. HC-2000 was sprayed in the drainage ditch to desorb additional oil from the soil and collected by several applications of sorbent worked into the soil. Polypropylene sorbent boom filtration barriers were placed in the drainage ditch to retain HC-2000 to maximize degradation retention times.

Oil covered metal supports for electrical equipment and concrete pads were scrubbed with HC-2000. Soil in and above the grounding grid zone were treated with a 6% solution of HC2. The treated area was covered with fresh gravel.

Residual oil in the grounding grid zone and drainage ditch were reduced to concentrations that allowed accelerated natural attenuation to continue to degrade via the slow release nutrients and catalysts in HC-2000.



Regulator Explosion at Substation



Removal of Free Product with Poly Sorbent



HC2 Application to Desorb Soil from Drainage Ditch



Recovery of Oil from Storm Drain and Desorbed Oil from Ditch Soil



Grounding Grid Soils and Metal and Concreted Pads Treated with HC2



Completed Substation Pad with Clean Gravel

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