

August 26, 1997

ENVIRONMENTAL
PROTECTION
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**REMEDIAL ACTION PLAN
625 HEGENBERGER BLVD. RD.
OAKLAND, CALIFORNIA
AEI PROJECT NO. 2233**

8/26/97

Prepared For:

**Diversified Investments Management Group
400 Oyster Point Blvd.
Suite 415
South San Francisco, California**

Prepared By:

**All Environmental, Inc.
111 N. Sepulveda Boulevard
Suite 250
Manhattan Beach, CA 90266
Phone 310-328-8878
Fax 310-798-2841**

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FIGURES

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APPENDIX B THE BIOREMEDIATION PROCESS

1.0 INTRODUCTION

This Remediation Action Plan (RAP) proposes to enhance the biodegradation of the petroleum hydrocarbons in the groundwater through the injection of microbes, oxygenated water, and nutrients into the groundwater. On behalf of Diversified Investments Management Group, the owners of the subject site, 625 Hegenberger Blvd. in Oakland, California, AEI is submitting this workplan to the Alameda County Health Services Agency for review and concurrence.

1.1 Site Background

The site, located at the corner of Hegenberger Road and Collins Drive in Oakland, California (See Figure 1), is a former fuel service station, which was developed in the mid 1960s. In the service station operated through the 1970's, when the tanks were abandoned. An automobile tune-up shop and convenience store have occupied the site until recently. The cinder block building, which once contained the convenience store and automobile repair shop, has been demolished. The subject site is currently unimproved.

In 1993 three 12,000 gallon USTs, six fuel dispenser pumps, and one sump were removed from the south portion of the subject site under the supervision of Levine Fricke (LF). Soil samples collected during the tank removal revealed that a significant hydrocarbon release had occurred. Soil sampling data from the tank removal and subsequent investigations indicated that there had been a release of gasoline, diesel, and waste-oil. The release had occurred beneath the tanks, from under the dispenser pumps, and from around the sump.

LF and Subsurface Consultants have drilled 24 shallow soil borings and LF has installed six groundwater monitoring wells. Samples collected from these borings and wells revealed significant soil and groundwater contamination. However, the extent of contamination has been defined.

In 1996 AEI remediated the contaminated soil. During the spring and summer of 1996, AEI excavated soil, which was considered a source of continued groundwater contamination. AEI excavated a total of 1,200 cubic yards (cy) of contaminated soil and combined that with the 300 cy of contaminated soil already stockpiled on the subject site. A total of 1,500 cy of soil was treated through on-site aeration and returned to the excavation. AEI documented this process in a final report dated March 3, 1997.

Quarterly monitoring of the six monitoring wells on-site was performed by LF through January, 1995. AEI began monitoring the wells in October of 1995 and has implemented an ongoing quarterly monitoring program. One of the six wells (MW-24) was destroyed in the spring of 1996 prior to beginning the soil remediation.

The last quarterly monitoring report was submitted to ACHCSA on May 27, 1997. This quarterly monitoring for June 17, 1997 revealed significant levels of hydrocarbons in wells MW8, MW11, and MW16.

The levels of hydrocarbon in the groundwater do not appear to be dropping and natural biodegradation appears to be minimal. Oxygen levels in the contaminated wells are significantly depleted and nitrate concentrations were below the detection limit. This explains the lack of microbial activity.

Mr. Barney Chan of ACHCSA replied to this quarterly monitoring report with a letter requesting a Remedial Action Plan (RAP) be prepared outlining plans to stimulate biodegradation. This RAP has been prepared in response to that request.

1.2 Project Objectives

This workplan is designed to meet the following objectives: 1) to enhance in-situ bioremediation by injecting microbes, oxygenated water, and nutrients; 2) to immediately reduce TPH gasoline and benzene concentrations to below action levels; 3) to provide the aquifer with increased capacity of on-going passive bioremediation.

AEI intends to provide the highest contaminant reduction possible within the budget of our proposal. But, as a minimum, the following levels will be achieved prior to terminating active bioremediation.

Compound	Proposed Cleanup Level
TPHg	1000 ug/L
TPHd	1000 ug/L
Benzene	100 ug/L

(probably OK)

post to cleanup
req. for future site
usage

Once these levels have been achieved, AEI will stop stimulating the metabolic bacteria population. However, the metabolic breakdown of petroleum compounds will continue, as the oxygen and nutrient levels will remain high for sometime.

Once the action levels have been achieved and confirmed through successive quarterly monitoring events, AEI believes this site should expeditiously be considered for closure. — OK

2.0 Evaluation of Remedial Alternatives

The remediation options for contaminated groundwater are to pump-and-treat, or to treat in-situ.

2.1 Pump & Treat

This method involves the pumping of the groundwater to remediation tanks, treating the groundwater, and discharging to the sewer or the subsurface.

This process is slow and has had very little success due to the fact that soil at the capillary fringe remains providing an on-going source for future groundwater contamination. Case history in California shows that very few sites have been closed using this method.

2.2 In-situ Bioremediation

Treating groundwater in place is possible by creating an environment in which bacteria, oxygen, and nutrients are available to degrade dissolved hydrocarbons in the groundwater. This process is known as bioremediation. Bioremediation is a natural process where microorganisms decompose the hydrocarbon contaminant into harmless fatty acids, carbon dioxide, and water. The attached article "The Bioremediation Process" gives a detailed ~~and~~ explanation of the process (Appendix B). It is a passive system that is safe and effective and substantially more cost effective than most other technologies.

In-situ bioremediation is the best available technology due to the contaminant biodegradability and ease of access into the aquifer. This is the recommended technology.

2.3 should evaluate as 3rd option
 No action, alternative.

3.0 REMEDIATION DETAILS

The mitigation at this site involves the injection of bioremediation enhancing inoculation into the groundwater through five new inoculation wells.

3.1 Inoculation Details

The site will be treated with an inoculation medium consisting of microbes, nutrients, time released oxygen, a biosurfactant type material, and water. All bioremediation products are manufactured by Ultra Coatings and have the trademark "UC-40". Product description, MSDS and safety information is shown in Appendix A. Each ingredient is discussed below.

- May not be
needed
- 11
- exp
- **Microbes:** the inoculate will consist of UC40-1014 microbes. These are a broad spectrum blend used on a wide range of hydrocarbons.
 - **Nutrients:** the UC-40 Microbe Nutrient is a precise blend of macro and micro nutrients designed to be used with the 1014 microbes.
 - **Oxygen:** a time released oxygenator called UC-40 O2TR will be used to maintain aerobic action. It maintains dissolved oxygen (DO) without drastic fluctuations in pH. It is a much more effective source of oxygen than mechanical aeration. Figures 2 and 3 show details of the system.
 - **Oil Breaker:** the UC-40 product enables water and petroleum molecules to merge thereby increasing the oil/water interface providing the bacteria a more accessible food source.

3.2 Distribution System Details

Inoculation into the groundwater will be done from five new injection wells installed * within the footprint of the contamination plume. *The excavation created during the soil remediation was backfill with 300 cy of peagravel, which will effectively serve as a sump during the injection of inoculate. The 300 cy of peagravel form a one foot layer just above the water table in the center of the plume. See Figure 3.

All five injection wells will have separate horizontal supply lines to the equipment area for individual feeding of inoculate and maintenance material. Each well will have a well cover, locking cap, and a sampling port. Figures 2, 3, and 4 show details of the wells and supply lines.

Weekly and monthly groundwater samples will be collected and analyzed by the schedule in Table 1. Adjustments to the inoculate will be made as required to maintain the optimum degradation rate.

3.3 Calculations of Inoculate Quantity

The quantity of each inoculate component will be based on the existing levels of nutrients and dissolved oxygen in the water. A sample will be taken from existing groundwater monitoring wells and analyzed for DO, nitrogen and Phosphorous. Based on those readings approximate quantities of nutrients and O2TR can be determined. Upon installation of the five inoculation wells individual samples will be taken and analyzed. Adjustments in the inoculant makeup will be made at that time.

will not
be done
as proposed

All water used for inoculation will be pumped from the center inoculation well. ~~500~~
Inoculation will be from the perimeter wells. Inoculation will be in ~~1,000~~-gallon batches with 24 hours difference between pumping and injection. This will result in a small hydraulic gradient to the center of the plume and have no net gain in groundwater quantity. This will minimize the possibility of "pushing the plume".

3.4 Agency Notification and Permits

The ACHCSA is the lead agency for this site and their approval will be sought by submitting this workplan for their review. Information as to the contents of the chemicals being injected into the ground is contained in Appendix A.

AEI will obtain a drilling permit form Alameda County Public Works Agency for the installation of five additional injection wells.

Tunnell.

3.5 Schedule

The speed of degradation is based on the concentrations of contaminant, types of contamination, and the optimization of the environment for the microbial process such as adequate oxygen, nutrients and temperature. Complex factors such as the variable aquifer conditions and the adequacy of the site characterization make it difficult to estimate the time required for closure, but experience and past ongoing projects show that closure levels may be reached within 6 months.

4.0 Monitoring & Closure

Monitoring the success of this process involves tracking significant factors in the soil and groundwater. Prior to the start of the groundwater treatment, the baseline data will be obtained to help assess the effectiveness of the remediation system. The following presents a discussion of the items to be monitored. A summary of the monitoring schedule is presented as Table 1.

TABLE 1					
TEST	BASELINE	WEEKLY @ Inoculation	MONTHLY	QUARTERLY	CLOSURE
Groundwater					
DO	YES	YES	YES	YES	YES
Nitrates	YES	YES	YES	YES	YES
TPH 8015M	YES	NO	YES*	YES	YES
Vapors					
CO2	YES	YES	YES	NO	NO

→ *microbe count? A*

* AEI will monitor on a monthly basis for the first month and then revert to a quarterly basis.

4.1 Groundwater Monitoring Wells

The wells at the perimeter of the plume will be monitored monthly during inoculation and quarterly thereafter. The purpose of the monitoring is two-fold. First the data will indicate that the contamination is being reduced, and secondly, that the contamination is not spreading beyond existing limits.

4.2 Dissolved Oxygen

The inoculation included the addition of oxygenated water. Each week the site will include sampling and testing for dissolved oxygen at the discharge point of the treatment tank. A record shall be kept of the DO readings and included in the monthly monitoring reports.

4.3 Carbon Dioxide

In-situ bioremediation is a process that gives off carbon dioxide. This RAP includes the installation of two vapor probes. Each month one sample shall be obtained from the site using standard gas sampling procedures. The carbon dioxide levels will increase as the microbes digest the plume and grow. Once the plumes has been significantly degraded carbon dioxide levels will decrease as the microbes die.

4.4 Bromide Ion

AEI will inject a non-reactive bromide ion which will serve as a tracer. The bromide ion will indicate the extent to which the inoculant ~~is~~ spread within the aquifer.

WAS


4.5 Closure Procedure

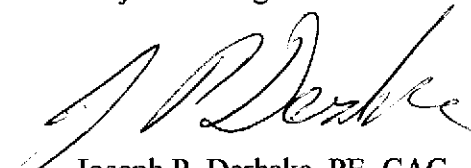
Closure groundwater samples will be collected from the three contaminated groundwater monitoring wells, MW-8, MW-11, and MW-16. Groundwater samples will be collected using the same sampling procedures, which have been applied in the quarterly groundwater sampling events.

5.0 Professional Signature

These services were performed in accordance with generally accepted practices in the environmental engineering and construction field at this time. This report was prepared under the responsible charge of Joseph P. Derhake, PE, a registered civil engineer in the State of California.

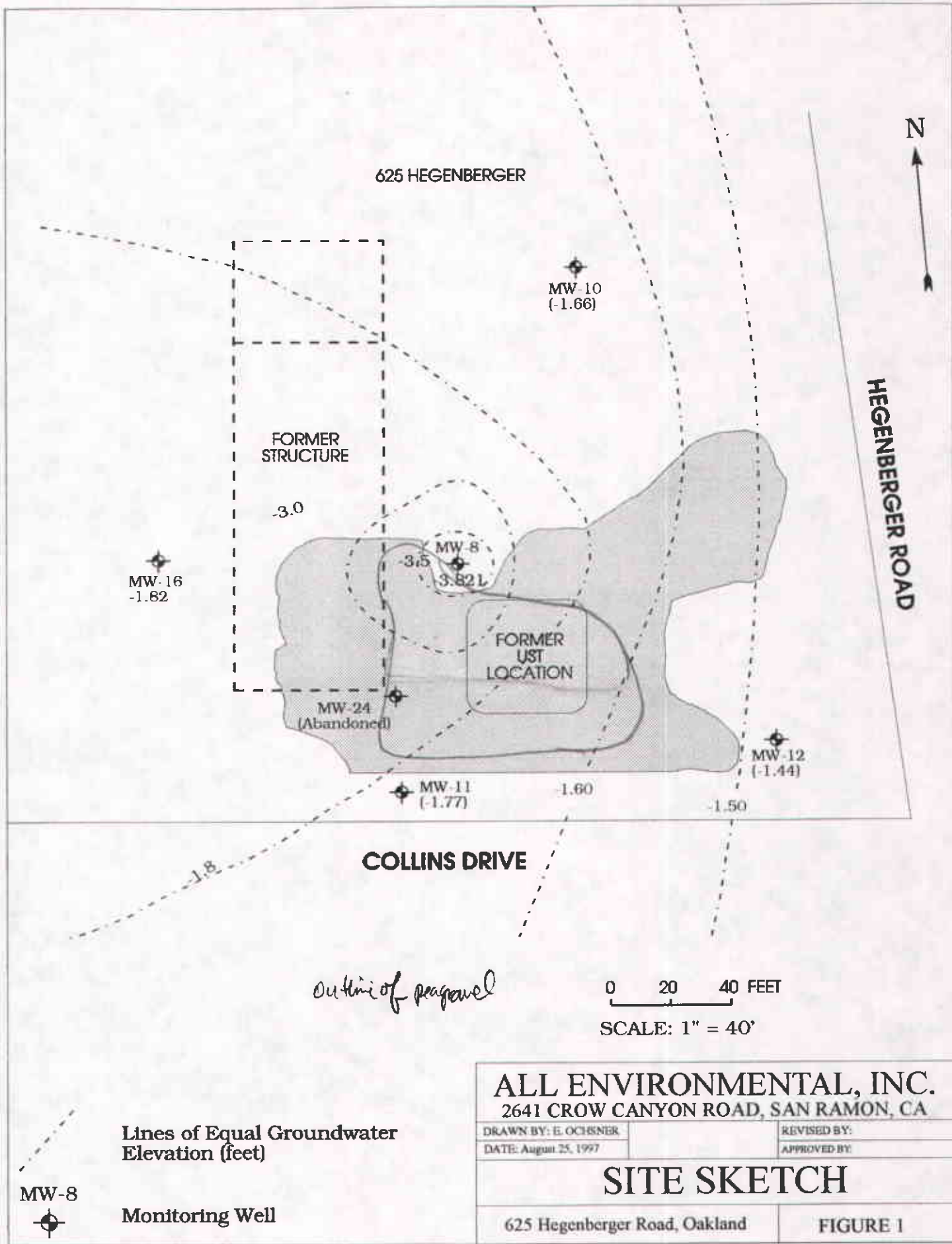
Sincerely,
All Environmental, Inc.


Bryan Campbell
Project Geologist


Joseph P. Derhake, PE, CAC
Project Manager







Inoculation Wells

Estimated Plume Limits

Supply Lines

Existing Monitoring Wells

Peagravel Location

EQUIPMENT
AREA

*need to sample
prior to closure*

HEGENBERGER ROAD

COLLINS DRIVE



ALL ENVIRONMENTAL, INC.

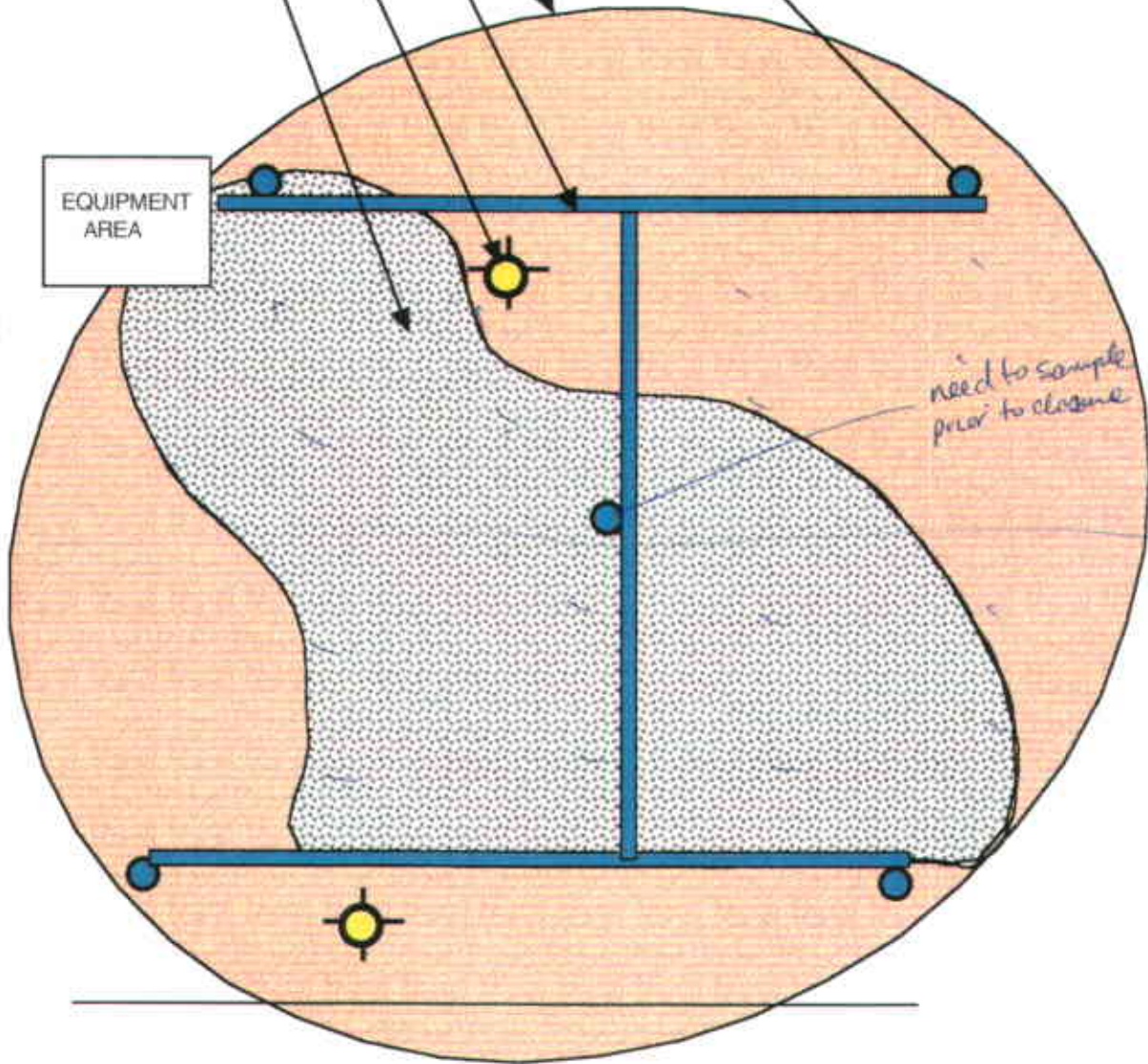
111 N. Sepulveda Boulevard, Suite 250, Manhattan Beach, CA

Scale: 1"=40'

Piping Layout-Plan View

625 Hegenberger Road,
Oakland, CA

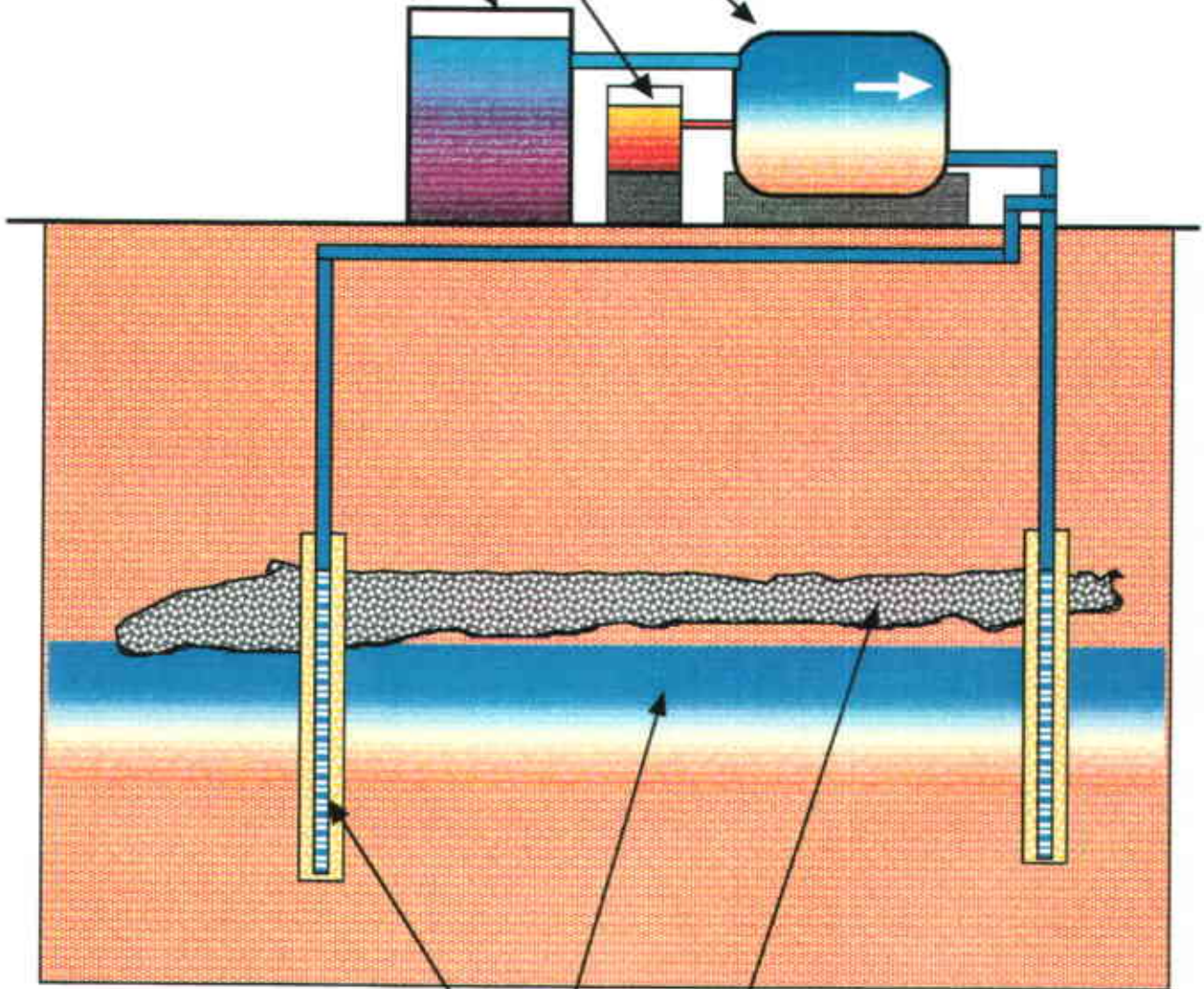
Figure 2



Bio Mixing Tank

Injection Pump

Biocatalyst Generator



Inoculation Wells

Contaminated Groundwater
and Saturated Soil

Peagavel sump



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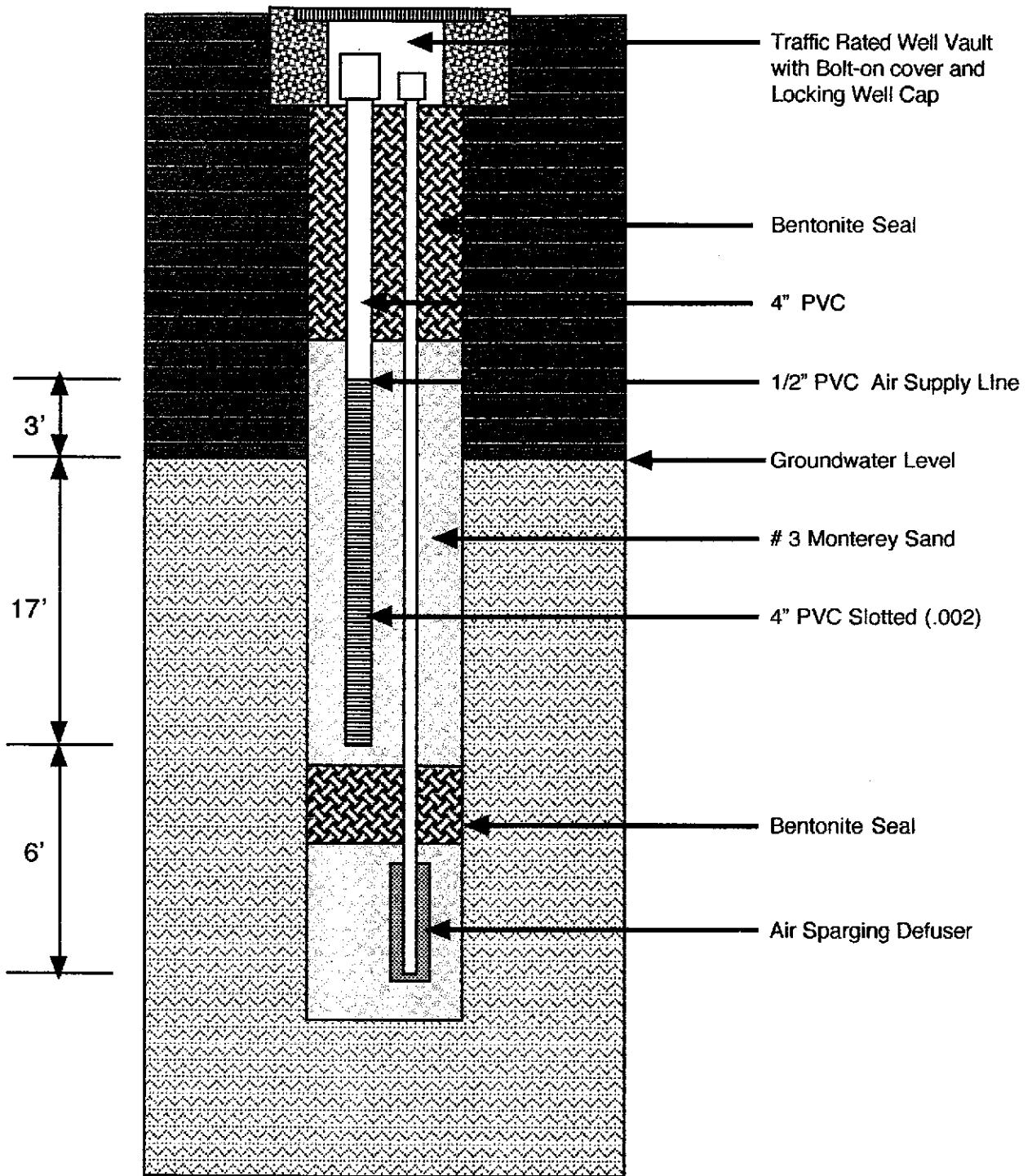
111 N. Sepulveda Boulevard, Suite 250, Manhattan Beach, CA

Scale: 1"=40'

Groundwater Treatment System

625 Hegenberger Road,
Oakland, CA

Figure 3

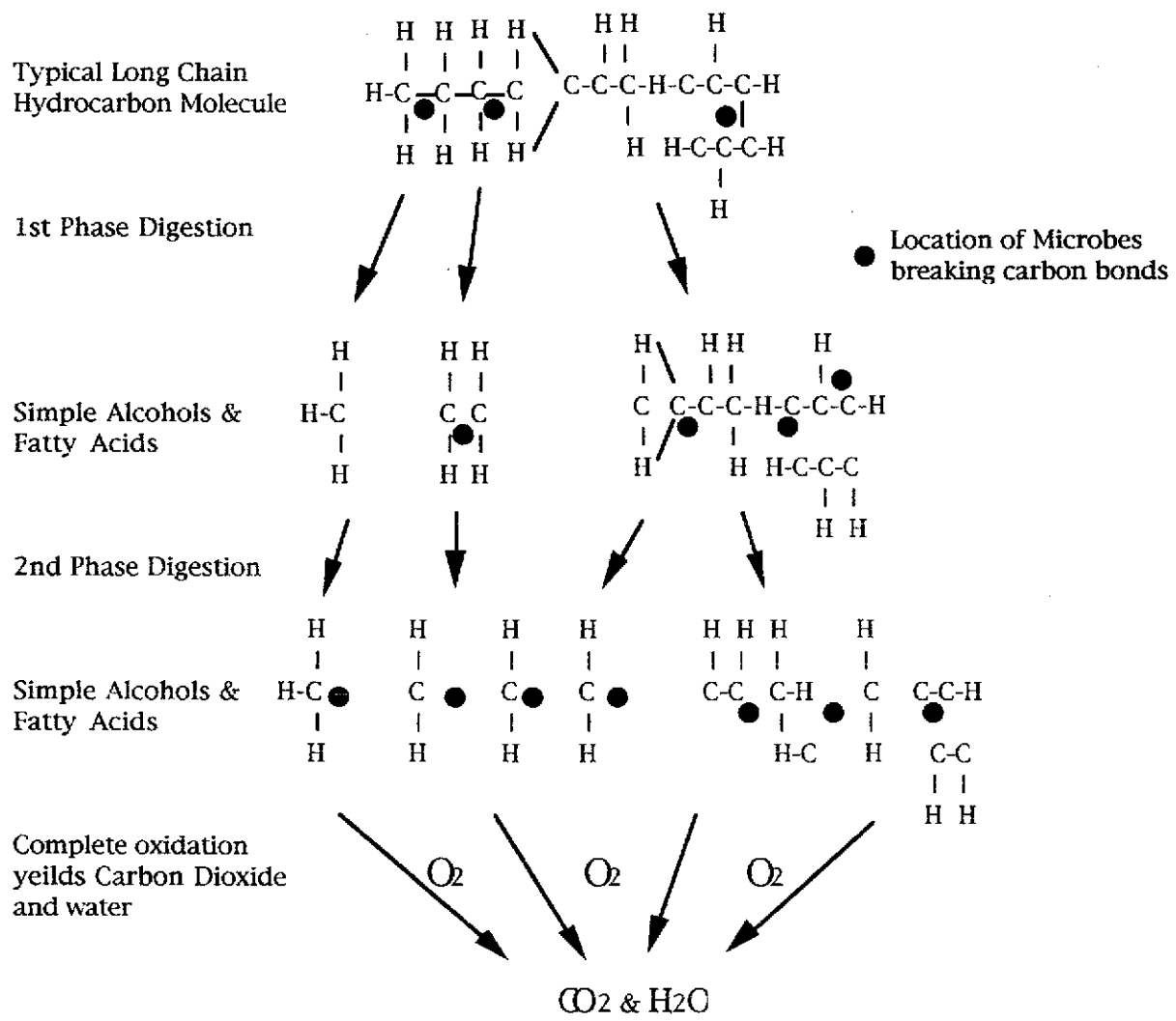


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111 N. Sepulveda Boulevard, Suite 250, Manhattan Beach, CA

Well Design

625 Hegenberger Road
Oakland, CA



SAMPLE DEGRADATION CHAIN

Bioremediation is the process of using natural microorganisms to recycle of break down organic materials. This diagram shows an example of the breakdown of a typical hydrocarbon molecule.

ALL ENVIRONMENTAL, INC.	
111 N. Sepulveda Boulevard, Suite 250, Manhattan Beach, CA	
Scale: 1"=40'	
Degradation Chain Diagram	
625 Hegenberger Road, Oakland, CA	Figure 5

APPENDIX A
MATERIAL SAFETY DATA SHEETS

UC-40™ Oil Breaker

microbial solution for petroleum hydrocarbon bioaugmentation

biodegradable water soluble nonflammable nonpathogenic

What

Bioremediation uses bacteria to consume hydrocarbons in soil, water, and on hard surfaces. Biostimulation uses bacteria already present in the matrix. Bioaugmentation uses a large volume of contaminant-specific bacteria from outside the matrix and places it where it is needed.

UC-40™ Oil Breaker is leading edge bioaugmentation technology. The distinct characteristics of this emulsion, in combination with specially blended bacterial consortia, revolutionizes bioaugmentation and makes biostimulation obsolete. Relying on bacteria already present in a matrix (biostimulation) is a time consuming and uncertain proposition.

How

Microbial degradation takes place at the oil/water interface. Oil Breaker's unique formula enables water and petroleum molecules to merge. This integration increases the oil/water interface and makes the hydrocarbons available to the bacteria as an immediate food source. This product accelerates what happens naturally over a longer period of time.

Features

This breakthrough technology improves the percolation rate of soil, adjusts matrix pH, and buffers substances toxic to bacteria. Oil Breaker creates ideal conditions for bacterial consumption of petroleum hydrocarbons, while ensuring the proper balance of micro and macro nutrients for cell construction and rapid reproduction. Petroleum hydrocarbon remediation is now fast, easy, safe, and economical.

Conditions

UC-40™ Oil Breaker and Microbe Formulas continue to work as long as:

- temperature is between 38-102° F
- pH is between 6.0-9.0
- water, oxygen, and nutrients are available
- there is a petroleum food source, e.g., oil, diesel, BTEX, Jet A, etc.

Directions

See Application Instructions for complete information on Solution Preparation, Equipment, Methods & Procedures for soil, water, & hard surface remediation.

Availability

4 gallon/case 55gallon/drum

MATERIAL SAFETY DATA SHEET

Identity (As Used on Label and List):

UC-40™ OIL BREAKER

DOT: Class Not Required

Formula: Proprietary

Section 1.

Supplier's Name
Ultra Coatings, Inc.
Address
1146 NW 52nd, Seattle WA 98107-5129

Emergency Telephone Number
1/800/424-9300
Telephone Number for Information
206/781-7117

Section 2. HAZARDOUS INGREDIENTS/IDENTITY INFORMATION

No Hazardous Ingredients

No toxic chemicals subject to the reporting requirements of Sec 313 of Title III and of 40 CFR 372 are present.

Section 3. PHYSICAL/CHEMICAL CHARACTERISTICS

Boiling Point	212°F	Specific Gravity (H ₂ O=1)	1.0	Vapor Density	heavier than air
Solubility in Water	complete	pH	7 (neutral)	Evaporation Rate	slower than ether
Appearance & Odor	pink liquid; semi-viscous, faint sweet odor				

Section 4. FIRE AND EXPLOSION HAZARD DATA

Flash Point (C.O.C.)	none	Flammable Limits in Air by Volume	Lower	n/a	Upper	n/a
Extinguishing Media	foam, CO ₂ , dry chemical, water fog					
Unusual Fire and Explosion Hazards	none					
Special Firefighting Procedures	Do not enter confined fire spaces without protective clothing & self-contained air supply.					

Section 5. REACTIVITY DATA

Stability	stable	Conditions to Avoid	none
Incompatibility (materials to avoid)	none	Hazardous Polymerization	will not occur
Hazardous Decomposition or Byproducts	May liberate carbon monoxide, carbon dioxide, and oxides of sulfur.		

Section 6. HEALTH HAZARD DATA

Routes of Entry	inhalation, ingestion, skin or eye contact		
Health Hazards, Acute and Chronic	none		
Conditions Aggravated by Exposure	none reported		
Carcinogenicity	NTP? no	IARC Monographs?	no
Emergency First Aid Procedures	Inhalation	Move to fresh air	
	Ingestion	If conscious, drink 2 glasses water. Consult physician.	
	Eye	Irrigate with water for at least 15 minutes, get medical attention	
	Skin	Water rinse for at least 15 min, remove contaminated clothing & shoes.	

Section 7. PRECAUTIONS FOR SAFE HANDLING AND USE

Precautions to be Taken in Handling	General good industrial practice is sufficient.
Steps to be Taken in Case Material is Released or Spilled	Wear suitable protective equipment. Small spills could be flushed with large quantities of water. Collect larger spills for disposal.
Waste Disposal Method	According to local, state & federal regulations.
Precautions to be Taken in Handling and Storing	Store in tightly closed container at temperature range of 40-120°F.
Other Precautions	This is an industrial product, not intended for home use. Keep away from children.

Section 8. CONTROL MEASURES

Respiratory Protection (Specify Type)	none required in good ventilation	Ventilation	normal
Protective Gloves	recommended	Local Exhaust	normal
Eye Protection	recommended		
Other Protective Clothing or Equipment	normal		
Work/Hygienic Practices	Exercise reasonable personal cleanliness. Wash hands after use.		

IT IS THE RESPONSIBILITY OF THE USER TO COMPLY WITH
ALL APPLICABLE FEDERAL, STATE AND LOCAL LAWS AND REGULATIONS.

NOTICE: The information presented herein is accurate to the best of our knowledge and is offered in good faith. No warranty, either expressed or implied, is hereby made. The recommended industrial hygiene and safe handling procedures are believed to be generally applicable. However, each user should review these recommendations in the specific context of the intended use and determine whether they are appropriate.

UC-40™ Microbe Formulas

These descriptions serve only as guidelines. Due to the high degree of variability of non-domestic waste streams, even within a given industry, conduct treatability studies to determine the preferred formulation. Ultra Coatings, Inc. can also customize formulations where it is warranted. For special instructions, submit a Project Data Sheet to your distributor for analysis.

Important

Do not combine microbe formulations. Doing so will kill the bacteria. Store in cool, dry place. Avoid heat and moisture at all times. Maintain temperature range of 45°-80°F. Do not store in deep freeze. Moisture activates product. Place in insulated cooler if temperature exceeds 100°F during storage or transportation.

Petroleum Hydrocarbons

1014 - Broad Spectrum

Multiple bacterial strains and sub-phyla make this formula capable of treating a wide range of hydrocarbons found in combination: crude, bunker C, creosote, coal tar, diesel, motor oil, gasoline, mineral spirits, etc. Self-regulating bio-surfactants are characteristic of this blend and dramatically aid heavy oil suspension for easy degradation. Useful where a variety of contaminants are known to be present.

1015 - Cold Weather, Broad Spectrum

Identical to the 1014 but able to maintain a rapid reproduction rate and consume more hydrocarbons at lower temperatures than other bacterial blends. Can be used in environments with ambient temperatures as low as 36 degrees F.

1030 - Chlorinated Solvents

The premier formula for chlorinated solvents. With proper application, the 1030 bacteria quickly and safely eliminate chlorinated solvents such as 1-1-1's, TCE, PCB's, and phenolics. Other biological systems rely on co-metabolites to induce artificial enzymatic action. The 1030 blend naturally breaks down hydrocarbon structures without hazardous additives. Valuable to industries where use and generation of such wastes is necessary and ongoing.

Non-Petroleum Hydrocarbons, FOGS: Fats, Oils, & Grease

1016 - Animal /Vegetable , Light Petroleum Oils

Remediates organic fats, oils, and greases. For waste streams with high lipid content from organic sources such as corn oil, lard and soybeans. Effective formula for aliphatic petroleum compounds, such as mineral spirits or Stoddard solvent. Useful in rendering plants, meat processing, or paint manufacturing where compounds like linseed oil are used during production.

continued

MATERIAL SAFETY DATA SHEET

Identity (As Used on Label and List):

UC-40™ MICROBE FORMULA

Identity: Microbial Cultures DOT: Class not required.
Formula: Proprietary Chemical Family: A mixture of soil microorganisms in combination with Calcium Carbonate. No hazardous components.

Section 1.

Supplier's Name

Ultra Coatings, Inc.

Address

1146 NW 52nd, Seattle WA 98107-5129

Emergency Telephone Number

1/800/424-9300

Telephone Number for Information

206/781-7117

Section 2. HAZARDOUS INGREDIENTS/IDENTITY INFORMATION

Biological Entity - Microbes (no hazardous ingredients)

Section 3. PHYSICAL/CHEMICAL CHARACTERISTICS

Boiling Point	none	Specific Gravity (H ₂ O=1)	n/a	Vapor Density (air=1)	1.2
Solubility in Water	insoluble/dispersible	pH of 10% solution	7.0±0.2	Flammable Limit	n/a
Moisture (% wet wt)	6%-12%	Vapor Pressure mm/Hg	n/a	Bulk Density	4.2-5.1 lb/gal
Appearance & Odor	tan, yeast-like granules, mild musky aroma			Particle Size	1.588 mm mesh

Section 4. FIRE AND EXPLOSION HAZARD DATA

Auto-Ignite Temperature	n/a	Unusual Fire and Explosion Hazards	none	Flash Point	n/a
Special Firefighting Procedure	none	Minimum Ignition Temperature	715F	Extinguishing Media	water

Section 5. REACTIVITY DATA

Stability	stable	Incompatible Substance	none known
Hazardous Polymerization	will not occur	Hazardous Decomposition	none

Section 6. HEALTH HAZARDS

Routes of Entry inhalation, ingestion, skin or eye contact
Health Hazards, Acute and Chronic none
Carcinogenicity none
Conditions Aggravated by Exposure Membrane irritation by dry powder. Avoid dusty or enclosed environments.
Emergency First Aid Procedures
INHALATION Move to fresh air, loosen collar, contact physician.
INGESTION Drink large quantities of water to dilute, call physician if condition persists.
SKIN OR EYE Wash or irrigate with water, seek medical attention.

Section 7. PRECAUTIONS FOR SAFE HANDLING AND USE

Precautions to be taken in handling No danger from handling packaged material. Ensure containers remain sealed.
Precautions to be taken in case of spill Small spills can be washed away with water. Product is biodegradable. Large spills, if uncontaminated, can be returned to container. Dry sweep for disposal.
Disposal Procedures According to local, state & federal regulations.
Storage Requirements Clean, dry, normal room temperature between 41-104F (5C-40C). Keep out of sunlight.
Other Precautions This is an industrial product, not intended for home use. Keep away from children.

Section 8. CONTROL MEASURES

Respiratory Protection (Specify Type) NIOSH approved particulate dust mask
Ventilation normal Local Exhaust normal Mechanical (General) normal
Protective Gloves recommended
Eye Protection safety glasses recommended
Other Protective Clothing or Equipment normal
Work/Hygienic Practices Exercise reasonable personal cleanliness. Wash hands after use.

Section 9. BIOLOGICAL HAZARD DATA

Microbial Formulation Product formulation consists of a range of naturally-occurring microorganisms which are known to be non-pathogenic to man or animals.
Other Product has been shown to be free of *Salmonella* using procedures outlined by AOAC and USDA.

IT IS THE RESPONSIBILITY OF THE USER TO COMPLY WITH ALL APPLICABLE FEDERAL, STATE AND LOCAL LAWS AND REGULATIONS.

NOTICE: The information presented herein is accurate to the best of our knowledge and is offered in good faith. No warranty, either expressed or implied, is hereby made. The recommended industrial hygiene and safe handling procedures are believed to be generally applicable. However, each user should review these recommendations in the specific context of the intended use and determine whether they are appropriate.

UC-40™ Microbe Nutrient

water soluble nutrients for microbial growth

UC-40™ Microbe Nutrient is a precise blend of highly water soluble macro and micronutrients necessary to biological life. While not a food source for hydrocarbon degrading bacteria, these nutrients provide necessary building blocks for cell structure and rapid reproduction of the UC-40™ Microbe Formulas.

Macronutrients

carbon
hydrogen
nitrogen
phosphorus
potassium
magnesium
sodium

Micronutrients

calcium
iron
copper
cobalt
molybdenum
manganese
zinc

Bacterial activity is diminished when nutrition is inadequate or out of balance. UC-40™ Microbe Nutrient is scientifically formulated in exact proportions for optimal cell growth, biosynthesis, and reproduction. Maintaining proper nitrogen, phosphorus and potassium (NPK) levels is critical to the success of any project. In nature, adequate nitrogen levels result in green leafy plants. Proper phosphorus levels promote plants that sprout fruit and flowers. Sufficient potassium levels are common in clay soils and lead to extensive root networks among indigenous plants.

To determine if nutrient levels are adequate, perform an NPK test on a sample from the matrix. Adjust nutrient levels according to distributor recommendations.

Directions

Nutrients are necessary for "conditioning" the matrix before microbes are introduced. Apply nutrient treatment 24 hours prior to applying the first microbial solution. If nutrient requirements are substantial, apply nutrient and microbial solutions separately. For example, apply nutrient treatments on Day 0 and Day 6. Apply microbial treatments on Days 1, 3, 9, 12 according to instructions. When combining nutrients directly to microbial solutions, do not exceed these ratios: 1 oz. of nutrients per 10 gallons of solution or 1# nutrients per 150 gallons of solution.

Availability

1# containers 25# pails

MATERIAL SAFETY DATA SHEET

Identity (As Used on Label and List):

UC-40™ MICROBE NUTRIENT

Section 1.

Supplier's Name

Ultra Coatings, Inc.

Address

1146 NW 52nd, Seattle WA 98107-5129

Emergency Telephone Number

1/800/424-9300

Telephone Number for Information

206/781-7117

Section 2. HAZARDOUS INGREDIENTS

Chemical Name/Common Name CAS #

none

Percent (optional)

TLV (source)

Section 3. PHYSICAL DATA

Boiling Point

n/a

Specific Gravity (H2O=1)

n/a

Vapor Density (air=1)

n

Solubility in Water

100%

Evaporation Rate (butyl acetate = 1)

n/a

Vapor Pressure (mm Hg)

n

Appearance and Odor

granular crystals, red color, no odor

Melting Point

n

Section 4. FIRE AND EXPLOSION HAZARD DATA

Flash Point

n/a

Flammable Limits

Lower

n/a

Upper

n/a

Extinguishing Media

none needed

Special Firefighting Procedures

none

Note: Product consists of 28% Mono and Diammonium phosphate, which are used in UL-approved ABC Fire Extinguishers

Unusual Fire and Explosion Hazards

none

Section 5. REACTIVITY DATA

Stability

stable

Incompatibility (Materials to Avoid)

none

Hazardous Decomposition or Byproducts

none

Hazardous Polymerization

will not occur

Section 6. HEALTH HAZARD DATA

Primary Routes of Exposure

n/a

Signs and Symptoms of Exposure

n/a

Medical Conditions Generally Aggravated by Exposure

n/a

HEALTH HAZARDS (acute and chronic)

n/a

CARCINOGENICITY: N/A

NTP?

IARC monographs?

OSHA regulated?

Emergency and First Aid Procedures

Upon accidental ingestion give patient ample water. Seek medical attention.

Section 7. PRECAUTIONS FOR SAFE HANDLING AND USE

Steps to be Taken in Case Material is Released or Spilled

Mop up or vacuum & place in container for re-use. No safety factors involved.

Waste Disposal Method

According to local, state & federal regulations.

Precautions to be Taken in Handling and Storing

none

Other Precautions

none

Section 8. CONTROL MEASURES

Respiratory Protection (Specify Type)

none required

Ventilation

not required

Local Exhaust

not required

Special n/a

Mechanical (General)

not required

Other n/a

Protective Gloves

not required

Eye Protection

not required

Other Protective Clothing or Equipment

no special requirements

Work/Hygienic Practices

no special requirements

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UC-40™ O₂TR

time release oxygenator for bioaugmentation

time release concentrate stable pH no by-products

What

Bioremediation requires oxygen in order to succeed. UC-40™ O₂TR releases oxygen in water without residual by-products. Aerobic action is the most efficient way for bacteria to degrade petroleum hydrocarbons. This proprietary blend keeps facultative anaerobes in an aerobic state for more effective remediation.

Features

Time release formula dissipates in 6-21 days.

O₂TR increases dissolved oxygen (DO) levels in aqueous environments unaffected by the physical injection of air. When used in conjunction with other UC-40™ bioaugmentation products, it maintains (DO) without drastic fluctuations of pH. O₂TR generates more oxygen and distributes it more evenly than mechanical aeration methods.

O₂TR delivers the (DO) necessary for bacteria to remain aerobic and degrade hydrocarbons rapidly. Even in sterile soil or oxygen deficient water, O₂TR will consistently meet the high oxygen demands of growing microbial populations.

Directions

Due to the potency of this product, contact your distributor for site specific quantities and application instructions.

Availability

1# containers 12# / case 100# / drum

IS THIS H₂O₂ ?

ORE - Mg₂O₂

Microbe Count

4 applications.

DO₂ - 6 ppm
N - 4-5 ppm

Jack Roberts: San Diego:

Susan Warner:

Water Board:

(Washed)
W012

MATERIAL SAFETY DATA SHEET

Identity (As Used on Label and List)

UC-40™ O₂ T R

Section 1. PRODUCT IDENTIFICATION

Supplier's Name

Ultra Coatings, Inc.

Address

1146 NW 52nd, Seattle WA 98107

Emergency Telephone Number

1/800/424-9300

Telephone Number for Information

206/781-7117

Gary Williams 2 Phd.

Section 2. COMPOSITION/INFORMATION ON INGREDIENTS

Material/Component	CAS #	percent
calcium peroxide	1305 79 9	55% minimum
calcium hydroxide	1305 62 0	45% maximum

Basically the same as ORC which is magnesium peroxide

Section 3. HAZARD IDENTIFICATION

Emergency Overview

Oxidizer

Contact with combustibles may cause fire. Under fire conditions product may decompose releasing oxygen that intensifies fire. Deluge container with water at safe distance or in protected area. Airborne dust may be irritating to eyes, nose, throat and lungs. No significant long term inhalation hazard; irritation usually subsides after exposure ceases.

Health Effects

Section 4. FIRST AID MEASURES

EYES

Immediately flush with large amounts of water for at least 15 min, lifting upper & lower lids intermittently. See an ophthalmologist.

SKIN

Wash with water. If irritation occurs & persists, obtain medical attention.

INHALATION

Remove subject to fresh air. If breathing discomfort occurs & persists, obtain medical attention.

INGESTION

If swallowed, drink plenty of water. Obtain medical attention.

NOTES TO PHYSICIAN Modest irritation is the only expected effect and should have no serious consequences except perhaps in the case of direct eye contact. Contaminated external surfaces should be flooded with water and direct eye contact deserves ophthalmologic evaluation. If ingested, gastrointestinal irritation but not caustic burns are to be expected; dilution with water indicated, as may be gastric evacuation via emesis or lavage if large doses or severe irritation is evident. Demulcents should be helpful. No systemic effects are expected though human toxicity data is sparse.

Section 5. FIRE FIGHTING MEASURES

Extinguishing Media

Deluge with plenty of water.

Special Firefighting Procedures

Use flooding quantities of water. Use water spray to keep fire exposed containers cool.

Degree of Fire & Explosion Hazard

Under fire conditions may decompose and release oxygen gas.

Hazardous Decomposition Products

Oxygen that supports combustion and calcium hydroxide.

Section 6. ACCIDENTAL RELEASE MEASURES

Procedure for Release or Spill

Confine spill and place into container, dilute with large quantity of water for disposal. Do not return product to original container. Runoff to sewer may create fire or explosion hazard. Do not flush powdered material to sewer.

Section 7. HANDLING AND STORAGE

Handling

Avoid contact by using personal protective equipment. Use respiratory protective equipment when release of airborne dust is expected. If compounded with organics or combustible materials be sure to exclude moisture.

Ventilation

Provide mechanical local exhaust ventilation to prevent release of dust into work environment. If ventilation is inadequate or not available, use dust respirator and eye protection.

Storage

Keep material dry. Store in a clean cool place. Do not store near or expose to heat sources: steam pipes, radiant heaters, hot air vents or welding sparks. Avoid contact with reducing agents. Reacts with moisture. Keep containers tightly closed when not in use.

Section 8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Control Measures

Provide mechanical local exhaust ventilation to prevent release of dust into work area. If release is expected use respiratory protection.

Recommended Personal Protective Equipment

Respiratory

approved dust respirator with full face piece

Gloves

general purpose rubber or neoprene

Special Clothing & Equipment

long sleeve shirt, impervious aprons or clothing

Footwear

general purpose rubber or neoprene

Section 9. PHYSICAL AND CHEMICAL PROPERTIES

Melting/Freezing Point	decomposes on heating	Boiling Point	n/a	Vapor Pressure	n/a
Room Temperature	white powder	Odor	none	Vapor Density (air=1)	n/a
Specific Gravity (H ₂ O=1)	approx 2.9 bulk density 65#/cu ft	% Volatiles	n/a	Evaporation Rate (butyl acetate=1)	n/a
Solubility in H ₂ O # by wt	slightly soluble	pH as is	n/a	pH 1% solution	slurry 12-13
Odor Threshold	n/a	Density (g/ml)	n/a	Partition Coefficient N-octanol/water	n/a
Flash Point	n/a	Autoignition Temperature	non combustible		
Oxidizing Properties	oxidizer	Solubility	no data		
Explosive Properties	decomposition at high temperatures releases oxygen.				
Flammable Limits (air)	upper n/a	lower	n/a		

Section 10. STABILITY AND REACTIVITY

Stability	Stable: decomposition could occur when exposed to heat or moisture.				
Hazardous Polymerization	will not occur.				
Conditions to Avoid	Heat, moisture: heavy metals and dirt can accelerate decomposition.				
Materials to Avoid	heavy metals, organics				
Incompatibility	Heat: decomposes @ 275C. Grinding mixtures with organics, heavy metals.				
Major Contaminants Contributing to Instability	Mixtures with polysulfide polymers may ignite; heat, moisture, reducing agents.				
Hazardous Decomposition Products	Oxygen that supports combustion.				
Sensitivity to Mechanical Impact	Oxidizable materials can be ignited by grinding and may become explosive.				
Sensitivity to Static Discharge	No data available.				

Section 11. TOXICOLOGICAL INFORMATION

Eye Contact	Severely irritating to unwashed eyes; minimally irritating to washed eyes.				
Skin Contact	non-irritating				
Acute Effects from Overexposure	Dust; irritating to eyes, nose, throat & lungs.				
Chronic Effects from Overexposure	No chronic problems on record. (Effects considered include sensitivities, carcinogenicity, teratogenicity, mutagenicity, synergistic products & any medical conditions generally recognized as being aggravated by exposure.)				

Section 12. ECOLOGICAL INFORMATION

Environmental Fate	AS indicated by chemical properties, oxygen is released into environment.				
Environmental Effects	Effect of low concentrations on aquatic life is not determined.				

Section 13. DISPOSAL CONSIDERATIONS

Waste Disposal Method	Dissolve in water to allow release of oxygen and dispose via a treatment system in accordance with governmental agencies' regulations. Contact appropriate regulatory agency prior to disposal.				
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Section 14. TRANSPORT INFORMATION

DOT Proper Shipping Name	calcium peroxide	IATA	calcium peroxide		
IMDG	calcium peroxide	DOT Classification	5.1 oxidizer		
DOT Labels	oxidizer	DOT Marking	calcium peroxide UN 1457		
DOT Placard	5.1 oxidizer	UN #	UN 1457		
Hazardous Substance/RQ	n/a	49 STCC #	49187717		
Precautions to Take in Transportation	Place spilled product in suitable container & wash residue with plenty of water.				
Other Shipping Information	Packing Group II.				

Section 15. REGULATORY INFORMATION

OSHA Exposure Limits	Substances	calcium peroxide	calcium hydroxide
OSHA PEL-TWA		n/a	5 mg/cu.m.
STEL		n/a	n/a
Ceiling		n/a	n/a
Skin Designation		n/a	n/a
ACGIH TLV-TWA		n/a	5 mg/cu.m.
STEL		n/a	n/a
Ceiling		n/a	n/a
Skin Designation		n/a	n/a
Target Organ Effects	eyes & respiratory passages		
Carcinogenic Potential	calcium peroxide & components		

Regulated by OSHA	no	Listed on NTP report	no	IARC Group 1, 2A, 2B	no
US EPA Requirements Release Reporting CERCLA (40 CFR 302)	calcium peroxide & components				
Listed Substances	no	RQ	n/a		
Category	n/a	RCRA Waste #	n/a		
Unlisted substances	yes				
RQ	100#	Characteristic	ignitability, corrosivity	RCRA Waste #	D001
SARA Title III Sec. 313 (40 CFR 372)			calcium peroxide, calcium hydroxide	Listed Toxic Chemical	D002
Inventory Reporting SARA Title III Sec. 311/312 (40 CFR 370)					no
Substances			calcium peroxide, calcium hydroxide		
Hazard Category			immediate (acute) health hazard, fire hazard		
Planning Threshold			10,000 lb		
Emergency Planning SARA Title III Sec. 302-303 (40 CFR 355)					
Listed Substances	no	RQ	n/a	Planning Threshold	n/a
US TSCA STATUS	listed				

Section 16. OTHER INFORMATION

Product Uses UC-40™ O₂ T R is a solid peroxygen chemical designed for environmental applications. The product provides controlled release of oxygen in-situ which permeates throughout the substrate. Depending on the application, it can release oxygen in a few hours to several weeks.

NFPA 704 Health 1 Flammability 0 Reactivity 1 Special Hazard oxy
 (Degree of Hazard: 0=no hazard, 4= severe hazard)

11.95

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SAFETY INFORMATION

UC-40™ Microbe Formulas

UC-40™ Microbe Formulas are designed blends of naturally occurring, saprophytic bacteria. These microbes are cultured from various locations in nature and are not genetically altered or engineered. They do not metabolize other living organisms and rely on dead, carbon based materials as a food source. They are non-pathogenic and will not transmit or cause disease in higher life forms.

"Microbe" is merely a convenient name for any of hundreds of thousands of species of microscopic organisms that flourish on earth; the most numerous are the ones we call bacteria. For two billion years, bacteria were the only life on earth. 'Only one microbe in a thousand is a pathogen - what we think of as a germ,' said Lenore Clesceri of Rensselaer Polytechnic Institute in Troy, New York. The rest, neither we nor the planet could live without. They make what we want, and they get rid of what we don't want. They are the workhorses of biotechnology."
From "Bacteria: Teaching Old Bugs New Tricks" by Thomas Y. Canby. National Geographic, V.184, No. 2, August 1993.

Strict quality control standards ensure that these formulations remain free of pathogens and contamination throughout the culture, collection, and packaging process. In the interest of public safety, purity is never compromised during any phase of manufacture or distribution.

Human contact with these organisms is not dangerous. There are no acute or chronic health hazards associated with their use. They are stable and non-reactive and do not cause hazardous polymerization or decomposition. It is recommended that reasonable handling practices be exercised with this or any foreign material, i.e. gloves and dust mask. Microbes come attached to bran. Protect exposed membranes (eyes, nose) from contact with the bran dust. Consult manufacturer's MSDS (Material Safety Data Sheet) for additional information on physical specifications, storage, and disposal.

With advanced notice and for a fee, Ultra Coatings, Inc. can provide documentation from a FDA/EPA certified lab that a given batch or lot number is free of *E. Coli*, *Salmonella ssp.*, *beta-hemolytic Streptococcus*, and *Staphylococcus Aureus*. 1.96

LABORATORY ANALYSIS

The bioassay and microbial analysis of UC-40™ Oil Breaker and Microbe Formulas conducted in July 1995 conform to E.P.A. Good Laboratory Practices set forth in the 40 CFR (163.80-1). Testing was performed by BRL, Redmond WA, a lab accredited by Washington State Dept. of Ecology (#C 073) and licensed by the U.S. Dept. of Agriculture (reg.# 91R043).

Static Acute Fish Toxicity Test

The bioassay on UC-40™ Oil Breaker was completed according to Washington Hazardous Waste Regulation (WAC 173-303) and followed procedure issued by the Washington State Dept. of Ecology "Biological Testing Methods: Part A. Static Acute Fish Toxicity Test" (DOE 80-12, revised June, 1991). Rainbow Trout (*Oncorhynchus mykiss*) were used as test organisms to determine mortality rates when exposed to samples of the product at 1:100, 1:500, and 1:1,000 v/v concentrations. Each concentration mixture was tested at 100 mg/L and 1,000 mg/L exposure levels. A waste material is designated as extremely hazardous waste (EHW) if, at a concentration of 100 mg/L, >10 out of 30 cumulative deaths occur within 96 hr. This material is said to have a lethal concentration (LC₅₀) of <100 mg/L at 95% confidence level. Similarly, a waste material is designated a dangerous waste if, at a concentration of 1,000 mg/L, >11 out of 30 cumulative deaths occur within 96 hr. This material is said to have an LC₅₀ of <1,000 mg/L at 90% confidence level.

Dilution	Mortality Rates	
	100 mg/L	1,000 mg/L
1:1,000	0%	3% (1 out of 30)
1:500	3% (1 out of 30)	7% (2 out of 30)
1:100	0%	10% (3 out of 30)

Conclusion: No significant effect on conductivity, pH, total hardness, and total alkalinity of the dilution water at increased concentrations of 1,000 mg/L. The tested material is said to have an LC₅₀ of greater than 1,000 mg/L and is not considered to be a hazardous or dangerous waste.

Microbial Analysis for Human Pathogens

UC-40™ Microbe Formulas were screened for human pathogens. API test strips were used during biochemical testing. The methods are outlined in "FDA Bacteriological Analytical Manual," 7th Edition, pgs. 17-21, 27-38, and 439-444, 1992. All tests were run in duplicate.

Conclusion: The microbes contained none of the human pathogens (*E.coli*, *Salmonella*, *Streptococcus*, or *Staphylococcus*). Attempts to get growth on enriched media for specific pathogens were negative at all dilutions.

continued

Acute Oral Rat Toxicity Study

The bioassay on UC-40™ Oil Breaker was completed according to Section 81-1 "Acute Oral Toxicity Study" of the Pesticide Assessment Guidelines, Subdivions F Hazard Evaluation: Human and Domestic Animals, pgs. 34-39, Nov. 1982. The purpose of this test was to evaluate the toxic characteristics of a substance to humans and animals by determining the median lethal dose (LD₅₀), its statistical limits, and slope using a single exposure and a 14-day post-exposure observation period. LD₅₀ oral is a statistically derived single dose of a substance that can be expected to cause death in 50% of the animals when administered orally. The LD₅₀ value is expressed in terms of weight of the test substance (g, mg) per unit weight of the test animal (e.g. mg/kg). Rats were dosed by gavage with 5,000 mg of the sample material per 1,000 grams of body weight.

Conclusion: No associated mortalities at the 5,000 mg/kg level. The sample is considered to be safe.

Survey for Hazardous Components

Survey completed in Jan. '96 by N.E.T. Testing Labs, Portland, OR. Analysis performed according to EPA test method 8260.

Conclusion: Non detect.

APPENDIX B
THE BIOREMEDIATION PROCESS



THE BIOREMEDIATION PROCESS A Practical Guideline

The following information is provided by BIOTREATMENT INC. (BTI) - *The Critter Company* as an overview of the science of bioremediation. The information presented is taken from our experiences and from information provided by Dr. Carl Oppenheimer, a microbiologist affiliated with BTI.

Bioremediation is based on the concept that nature provides a mechanism for naturally recycling all organic material. Applied microbial bioremediation helps nature along by isolating specific organic degrading microbes, cultivating a large quantity and introducing them to the contaminated medium with proper amounts of nutrients and oxygen to dramatically accelerate the degradation process.

Applied microbial bioremediation can use either naturally occurring microbes or genetically engineered microbes (GEM's) or pathogens. **BTI and this article only deals only with naturally occurring microbes.**

The Microbe:

Bacteria are the earth's primitive single celled organisms. Their basic role is to recycle the components of living organisms, converting them to the nutrient chemicals used by plants in photosynthesis and chemosynthesis. The bacteria have an average size of one micron, a 10,000th of a centimeter or 25,000th of an inch. More than 3000 species of bacteria have been identified and many more than that are still unknown.

Their size makes the microbe one of the smallest living units that contain the necessary complex chemicals for life processes and the necessary enzymes for their role in recycling complex organic matter. These small cells, by design, have a large surface to volume ratio which permits a maximum cell wall chemical activity and interchange of materials into and out of the cells.

Each molecule that is produced by life is decomposed either during metabolism of higher organisms including plants, or recycled by microorganisms. The goal of nature's recycling is to release their elements back to the inorganic components, to be utilized again by plants and animals.

The design of our earth and living forms has a balance, or equilibrium, dictated by the laws of physics and chemistry, geological structures and the composition of plants and animals. The microbe fits into this balance by its small size, great tolerance for variations of temperature, water availability, concentration of materials and the necessary enzymes to recycle all the 6 million chemical organic compounds produced as a part of life.

Distribution in Nature:

Since the primary responsibility of microorganisms is to recycle organic material, and since the total biological protoplasm is relatively constant for any ecological system, bacteria of a wide variety of species must be present in sufficient quantities and diversity in all environments to recycle the organic material, both natural and man made.



BIOTREATMENT INC.

The Critter Company

Bacteria, because of their small size are readily distributed through out the earth's surface. They are transferred by wind into the atmosphere to heights of 80,000 feet in dust, in water currents carry microbes into the deepest ocean channels. Microorganisms have been found at the base of the deepest oil wells. It is estimated that an adult human may have as many as 3 pounds of bacteria on the skin and internal organs. These organisms continuously add to the surrounding environment.

Microorganisms have many other properties that characterize their role as mineralizers. They have the ability to form resting cells in times when food is not available. When environmental conditions become favorable, these resting cells can rapidly infiltrate the environment to fulfill their basic role in recycling organic matter. In soils, microorganisms are readily transported by water movement over long distances both down and through geological formations. In fact much of the weathering of rocks and soils are carried out by microbial activities. Oil, gas and coal are the products of living organisms trapped in geological formations.

The patterns of the distribution of soil and water microorganisms are more complex than stated above. However one can realize that nature developed a tremendous capability of producing and continually distributing a large population of these small very active and mobile single celled organisms. The Microbes have the basic responsibility to restore the environment wherever natural or man made pollutants are in excess. When pollutants are in excess of natural microbial recycling, the bacteria will continue to work but take longer than man is willing to wait. For this state of unbalance, man has coined the word pollution.

Bioremediation:

Applied Microbial Bioremediation is a relatively new term used to describe the enhanced recycling of human, industrial and agricultural wastes. The natural process can be accelerated by the application of selected microbial populations designed to supplement the natural microorganisms and thus direct their activity.

Applied microbial bioremediation therefore is a process where massive numbers of selected microorganisms are introduced into contaminated soil or water. The organisms are carefully selected for their ability to degrade contaminated materials to harmless by products and to use this process for growth and energy. These microorganisms will supplement and associate with indigenous microorganisms and through the proper application, the versatile mixture can materially enhance the normal cycles present in the environment.

Bioremediation with such specially selected mixtures of microorganisms can be used in- situ, in special reactors, on surface water or soil. The application methodology is dictated by the conditions of the general ecology of the area and the chemistry and concentrations of the contaminating material to be recycled.

Application:

Obviously, because of the many complex interrelationships between microorganisms and their environment, each site requires a separate approach. The primary object is to bring an inoculate of specific microorganisms, water, oxygen and nutrients into contact with the contaminated material.



For microbial enzymes to be functional there must be a contact between the microbial cell and the hydrocarbon molecule. The bioengineering of any site selected must include a thorough site analysis by a field ecologist and microbiologist. This should include the following information:

- Site Description
- Soil Information - NPK, pH ect.
- Soil Temperature
- Contamination Limits
- Required Treatment Levels
- Geological Information
- Soil type and porosity
- Water Table Level & Gradient
- Contamination Levels
- Toxic Materials in Soil

In above ground or shallow polluted areas the site analyses is quite simple. It becomes more complicated with depth and presence of a water table or underground aquifer flow.

Requirements for Effective Bioremediation

1. **Contact:** The microorganisms and hydrocarbon molecule must be in contact. The enzymes necessary to break down the large molecules must be excreted by the organisms thru the cell surfaces. As smaller by-products are produced they may be absorbed through the cell wall where internal enzyme reaction takes place thus continuing the degradation process.
2. **Nutrients:** Hydrocarbon oxidizing microorganisms and other degrading cells require inorganic nitrogen, phosphorous, potassium plus trace minerals. These nutrients must be available for optimal growth. Trace minerals, such as iron zinc and manganese are generally assumed to be present in the soil or water. The dominant nutrient is nitrogen whereas phosphorous and potassium can be added in excess.
3. **Water** is an essential part of all biological activity. Water can control bioremediation effects by limiting or enhancing nutrient distribution and oxygen transport. Saturated soils with organic matter including hydrocarbons soon become anaerobic. Fine sediments such as clay may have a large proportion of bound water which will reduced nutrient and oxygen diffusion. A scientific knowledge of the properties of soil and water is essential for proper bioengineering.
4. **Oxygen** is essential for aerobic metabolism. Without oxygen the microorganisms can not efficiently degrade the molecules. The geological evidence for the accumulation of oil, coal, humus and lignites in anaerobic sediments illustrates the protective action of an anaerobic system. Oxygen then is one of the most important parameters in applied bioremediation. There is a mechanism for oxygen production in-situ through the use of a biocatalyst which provides atomic oxygen.

Biocatalyst:

A catalyst is a compound which in small amounts accelerates a chemical reaction without being consumed in the process. A biocatalyst is a substance that initiates or modifies the rate of a biological process and is generally consumed in the process.

The biocatalyst is derived completely from natural materials and has been shown in many different applications throughout the years to stimulate biological activity. It is Dr. Oppenheimer hypotheses that oxygen can be catalytically transferred from the water molecule to atomic oxygen on surfaces at which time the oxygen would be almost instantaneously oxidized or be available at cellular surfaces. This reaction could take place at the atomic level, at the interface between microorganisms and surfaces. This atomic oxygen is very difficult to measure. Evidence of activity is generally determined by an increase in biological activity.

In the presence of the biocatalyst, microbial populations exceed those produced by normal oxygenation and nutrients. Laboratory and field test show a thousandfold increase in cell mass of microorganisms cultivated in the presence of the biocatalyst. In addition there is laboratory evidence that the biocatalyst accelerates growth in minimal oxygenated media. Aerobic growth can proceed at a more rapid rate suggesting that oxygen is available.

The biocatalyst in sediments could produce micro amounts of available atomic oxygen even under anaerobic conditions. This effect may be responsible for the successful use of facultative aerobes in microbial enhanced oil recovery where our microorganisms with catalyst was effective in increasing the yield of many low producing oil wells.

The use of an oxygen producing biocatalyst opens a new era of applied bioremediation in ground waters and soil where oxygen is at a minimum.

Toxicity:

Toxic materials include inorganic or organic compounds that will kill or inhibit a part or all the microorganisms. The complexity of the environment requires a compatibility test for each application.

Temperature:

Microorganisms are active in temperatures from freezing to 70 C. Normally activity is closely related to temperature increase. There is evidence that biological activity is stimulated by infra red radiation. In some cold environments it is possible to increase the biological activity by modifying a greenhouse effect.

Summary:

The rate of bioremediation will be relative to the concentration of hydrocarbons in the original site and the ability to provide optimal environmental conditions for microbial growth. Applied bioremediation does not consist only of the addition of microorganisms or the addition only of nutrients. The total environmental system, as related to the size and properties of the microbial amendment, must be considered. If any of the four basic requirements listed above are missing, then the process will not be efficient or may not take place. In addition, other properties such as soil types, soil chemistry, organic content, toxic materials, temperature, etc., are also important parameters, and as such must also be considered. Bioremediation is an efficient and cost effective way to treat contaminated soils and groundwater. The key is to understand the microbiology and geology of the site and design an inoculant and distribution system to account for those conditions.

MICROBIAL BIOCATALYST

The Microbial Biocatalyst manufactured and distributed by BIOTREATMENT INC. is derived from a process used by Dr. Carl Oppenheimer to create an oxygenated water that dramatically increased the microbial activity in soil and groundwater. The following presents a discussion of the biocatalyst taken from an article written by Dr. Carl Oppenheimer in June 1992 entitled "Applied Microbial Bioremediation, A Practical Guideline".

"A catalyst is a compound which in small amounts accelerates a chemical reaction without being consumed in the process. A biocatalyst is a substance that initiates or modifies the rate of a biological process and is generally consumed in the process.

Our biocatalyst is derived completely from natural materials and has been shown in many different applications throughout the years to stimulate biological activity. There are certain observations, during the use of this biocatalyst, that suggests it is interrelated with the availability of oxygen. We are familiar with the uptake of hydrogen and the catalytic influence of metals such as iron on the conversion of molecular hydrogen to atomic hydrogen. This is the basis for cathodic protection of iron. Is it possible that oxygen can be catalytically transferred from the water molecule to atomic oxygen on surfaces at which time the oxygen would be almost instantaneously oxidized or be available at cellular surfaces. This reaction could take place at the atomic level, at the microscopic interfaces between microorganisms and surfaces. This atomic oxygen is very difficult to measure. Evidence of activity is generally determined by an increase in biological activity.

In the presence of the biocatalyst, microbial populations exceed those produced by normal oxygenation and nutrients. It is possible that because of the physics of normal mechanical aeration (bubbles) in water, there is a microsphere of area between the bubbles where dissolved oxygen is very low. If so this could account for the thousand fold increase in cell mass of microorganisms cultivated in the presence of the biocatalyst. In addition there is laboratory evidence that the biocatalyst accelerates growth in minimal oxygenated media. Aerobic growth can proceed at a more rapid rate suggesting that oxygen is available. Thus a facultative anaerobic culture will continue to function as an aerobe even with the minimal concentration of molecular oxygen.

The biocatalyst in sediments could produce micro amounts of available atomic oxygen even under anaerobic conditions. This effect may be responsible for the successful use of facultative aerobes in microbial enhanced oil recovery where our microorganisms with catalyst was effective in increasing the yield of many low producing oil wells.

The use of an oxygen producing biocatalyst opens a new era of applied bioremediation in ground waters and soil where oxygen is at a minimum."

The Microbial Biocatalyst produced by BIOTREATMENT INC. is safe and effective. Both lab and field test have shown a dramatic increase in microbial activity due to the oxygen enhancement properties of our Microbial Biocatalyst.