

An Overview of the Phytoremediation of Lead and Mercury

Prepared By

Jeanna R. Henry

National Network of Environmental Management Studies (NNEMS)
Fellow

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Foreword

The potential use of plants to remediate contaminated soil and groundwater has recently received a great deal of interest. EPA's Technology Innovation Office (TIO) provided a grant through the National Network for Environmental Management Studies (NNEMS) to assess the status of the use of phytoremediation to clean up lead (Pb) and mercury (Hg) contaminated soil. This report was prepared by an undergraduate student from Salisbury State University during the summer of 2000.

About the National Network for Environmental Management Studies (NNEMS)

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

Each participating headquarters or regional office develops and sponsors projects for student research. The projects are narrow in scope to allow the student to complete the research by working full-time during the summer or part-time during the school year. Research fellowships are available in Environmental Policy, Regulations, and Law; Environmental Management and Administration; Environmental Science; Public Relations and Communications; and Computer Programming and Development.

NNEMS fellows receive a stipend determined by the student's level of education and the duration of the research project. Fellowships are offered to undergraduate and graduate students. Students must meet certain eligibility requirements.

Acknowledgments

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Table of Contents	Page
Purpose	-1-
1.0 Introduction	-2-
2.0 The Problem	-3-
2.1 Heavy Metals	-3-
2.1.1 Lead	-4-
2.1.2 Mercury	-5-
3.0 Overview of Phytoremediation	-9-
3.1 What is Phytoremediation	-9-
3.2 Advantages of Phytoremediation	-10-
3.3 Disadvantages and Limitations of Phytoremediation	-12-
3.4 Current Status and Projected Market of Phytoremediation	-13-
3.5 Environmental Concerns Associated with Phytoremediation	-14-
3.5.1 Ecological Risk Assessment	-15-
4.0 Mechanisms Used for the Phytoremediation of Heavy Metals	-18-
4.1 Phytoextraction	-18-
4.2 Phytostabilization	-20-
4.3 Rhizofiltration	-21-
4.4 Phytovolatilization	-22-
5.0 Overview of the Phytoremediation of Lead	-23-
5.1 Lead and the Soil Matrix	-23-
5.2 Phytoextraction of Lead	-24-
5.3 Role of Synthetic Chelates in Phytoremediation	-24-
5.4 Green House Study of Phytoextraction Using Synthetic Chelates	-26-
6.0 Summary of Recent Field Applications Involving Lead	-29-
6.1 <u>Bayonne, New Jersey</u>	-29-
6.2 <u>Dorchester, Maine</u>	-35-
6.3 <u>Trenton, New Jersey (Magic Marker Site)</u>	-36-
6.4 <u>Twin Cities Army Ammunition Plant (TCAAP); Site C and Site 129-3</u>	-37-
6.5 <u>Open Burn/Open Detonation Area at the Ensign-Bickford Company</u>	-38-
6.6 <u>Confidential Sites</u>	-40-
6.6.1 Superfund Site	-40-
6.6.2 Dump Site for Lead Acid Batteries	-41-
7.0 Overview of the Phytoremediation of Mercury	-43-
7.1 Mercury and the Soil Matrix	-43-

Overview of the Phytoremediation of Lead and Mercury

Jeanna R. Henry

7.2 Phytoremediation of Mercury	-44-
7.2.1 Transgenic Plants	-44-
7.2.2 Phytoextraction of Mercury	-45-
7.3 Bench Scale Study: Argonne National Laboratory West, Waste Area Group 9, Operable Unit 9-04 (ANL-W)	-45-

8.0 Conclusion	-47-
-----------------------------	------

References	-49-
-------------------------	------

Figures:

Figure 1 - Transport and Distribution of Mercury in the Environment	-6-
Figure 2 - Plant Processes Leading to Environmental Remediation	-9-
Figure 3 - Phytoextraction of Ni	-19-
Figure 4 - Engineered Rhizofiltration System	-22-
Figure 5 - Enhancement of Plant uptake of Pb through the use of Synthetic Chelates	-25-

Tables:

Table 1 - Substances Amenable to the Phytoremediation Process	-11-
Table 2 - Advantages and Disadvantages/Limitations of the Phytoremediation Process	-12-
Table 3 - Calculations of Ecological Quotients	-17-
Table 4 - Summary of Recent Field Applications Involving Lead	-31-

Overview of the Phytoremediation of Lead and Mercury

Jeanna R. Henry

Purpose

The purpose of this report is to assess the current state of phytoremediation as an innovative technology and to discuss its usefulness and potential in the remediation of lead and mercury contaminated soils found at hazardous waste sites. An overview of phytoremediation is provided and discusses the advantages and disadvantages/limitations, current status and projected market and environmental concerns associated with this new and innovative technology. A brief description of the technologies used for the phytoremediation of heavy metals follows, leading into the phytoremediation of lead and mercury contaminated soils. Case studies involving the phytoremediation of lead and mercury detailing bench and full-scale projects are also provided.

1.0 Introduction

Since the dawn of the Industrial Revolution, mankind has been introducing numerous hazardous compounds into the environment at an exponential rate. These hazardous pollutants consist of a variety of organic compounds and heavy metals, which pose serious risks to human health. Heavy metals are primarily a concern because they cannot be destroyed by degradation. Frequently, the remediation of contaminated soils, groundwater, and surface water requires the removal of toxic metals from contaminated areas.

2.0 The Problem

According to a 1997 report it is estimated that there are almost half a million contaminated sites throughout the United States and more than 217,000 of them are still in need of remediation [26]. The national clean-up market consists of the Environmental Protection Agency's (EPA) Superfund sites and RCRA, Department of Defense (DOD), Department of Energy (DOE), State sites, and Private Party sites. Superfund sites are the most contaminated hazardous waste sites located in the United States and are on the National Priorities List (NPL). RCRA, the Resource Conservation and Recovery Act, regulates hazardous waste treatment, storage, and disposal facilities [22].

Sixty-four percent of Superfund and RCRA sites are contaminated with both organic and heavy metal species and another 15% are contaminated solely by metals. Eleven percent of the DOD's 7313 sites, covering 26,000 acres, are contaminated with heavy metals. The DOE has 4,000 sites, 23 of them listed as Superfund sites, with 53% contaminated with organic compounds and heavy metals and 7% with metals alone. There are 19,000 state-owned sites with 38% containing heavy metals and organics and 7% with only metals. The number of Private sites in need of remedial action has been estimated at 24,000 [22].

2.1 Heavy Metals

The most common heavy metals at hazardous waste sites are Cadmium (Cd), Chromium (Cr), Copper (Cu), Lead (Pb), Mercury (Hg), Nickel (Ni) and Zinc (Zn) [26]. Of these, lead and mercury are two of the most significant contaminants, posing serious and sometimes life threatening health hazards. Lead, which contaminates more than 50% of sites found on the NPL, is one of the most prominent metal contaminants found in hazardous waste sites [29]. Mercury

also poses significant environmental and health concerns. The World Health Organization (WHO) has approximated that each year 10,000 tons of mercury are released globally from both natural and anthropogenic sources [15].

2.1.1 Lead

Sources of Lead

Lead (Pb) is a bluish-grey metal that occurs naturally in minute amounts within the Earth's crust. It has also been referred to as plumbum, lead metal, and pigment metal [11]. Frequent use in many industrial processes is the main reason for lead contamination of the environment. There are a variety of industrial processes that involve the use of lead such as mining, smelting, manufacture of pesticides and fertilizers, dumping of municipal sewage and the burning of fossil fuels that contain a lead additive. Many commercial products and materials also contain lead including paints, ceramic glazes, television glass, ammunition, batteries, medical equipment (i.e., x-ray shields, fetal monitors), and electrical equipment. The uses of lead for roofing and the production of ammunition has increased from previous years [11]. Lead battery recycling sites, of which 29 have been labeled Superfund sites, and manufacturers use more than 80% of the lead produced in the United States. On average, recycled lead products only satisfy half of the nation's lead requirements [14].

Forms of Lead

Ionic lead (Pb^{2+}), lead oxides and hydroxides and lead-metal oxyanion complexes are the general forms of lead that are released into the soil, groundwater and surface waters. The most stable forms of lead are Pb^{2+} and lead-hydroxy complexes. Pb^{2+} is the most common and reactive form of lead, forming mononuclear and polynuclear oxides and hydroxides [13].

The predominant **insoluble lead** compounds are **lead phosphates, lead carbonates** (form when the pH is above 6) and **lead (hydr)oxides** [22]. **Lead sulfide (PbS)** is the most stable solid form within the soil matrix and forms under reducing conditions when increased concentrations of sulfide are present. Under anaerobic conditions a volatile organolead (tetramethyl lead) can be formed due to microbial alkylation [13].

Health Effects

Lead has been listed as a **potential carcinogen** in the EPA Toxic Release Inventory (TRI) [11]. Inhalation and ingestion are the two routes of exposure, and the effects from both are the same. Pb accumulates in the body organs (i.e., brain), which may lead to poisoning (plumbism) or even death. The gastrointestinal tract, kidneys, and central nervous system are also affected by the presence of lead. Children exposed to lead are at risk for impaired development, lower IQ, shortened attention span, hyperactivity, and mental deterioration, with children under the age of six being at a more substantial risk. Adults usually experience decreased reaction time, loss of memory, nausea, insomnia, anorexia, and weakness of the joints when exposed to lead [11].

2.1.2 Mercury

Sources of Mercury

Mercury (Hg), also a naturally-occurring element is a silver-white liquid at room temperature. Due to this property, it is also referred to as **kwik, liquid silver, hydrargyrum**, and **metallic mercury**. The most common mineral form of mercury is the **non-toxic, insoluble mercuric sulfide or cinnabar (HgS)** a by-product obtained by the processing of complex ores that contain mixed sulfides, oxides, and chloride minerals [17]. Naturally occurring Hg is released by degassing of the earth's crust, volcanoes and the evaporation from oceans [3].

Mercury has a wide variety of uses in industry: medicine, dentistry, batteries, science, and military applications [12]. The burning of fossil fuels and medical waste incineration accounts for more than 80% of all anthropogenic sources [22]. Fifty-five percent of the total consumption of mercury is by chloralkali synthesis (used in electrodes), the wood pulping industry, paint, and electrical equipment. It has been estimated that the global reservoir of atmospheric mercury has increased by a factor of 2 to 5 since the beginning of the industrial revolution [3]. Atmospheric contamination by industry has recently decreased, but mining is still a significant contributor to

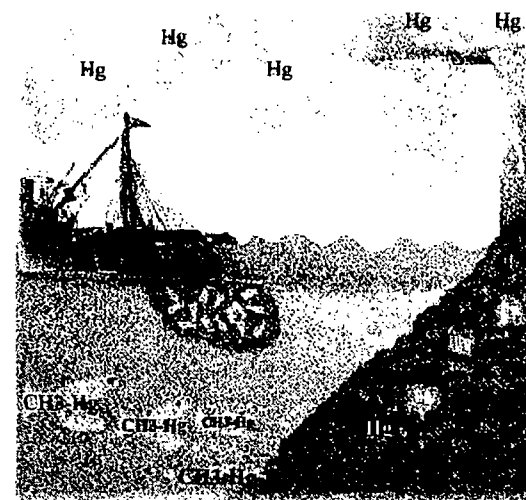


Fig 1: Transport and Distribution of Mercury within the Environment
Source: Measuring Mercury
[http://ehpnet1.niehs.nih.gov/docs/1996/104\(8\)/focus.html](http://ehpnet1.niehs.nih.gov/docs/1996/104(8)/focus.html)

the contamination of ground and surface waters. The smelting of lead, copper, and zinc ores emits approximately 100 tons globally and 9 tons throughout the US into the atmosphere on an annual basis [3].

Forms of Mercury

Mercury is transported and distributed in the environment through two processes. The first involves the atmospheric circulation of elemental mercury from land and water sources, which has a global effect [3]. Elemental mercury is initially released into the atmosphere, captured by precipitation and ultimately deposited in the sediments of lakes and oceans. This process leads to the second type of the transport and distribution of mercury. It involves the deposition of mercury in the sediments of lakes and oceans and its transformation to a methylated species by anaerobic bacteria. The amount of methyl-mercury produced by anaerobic bacteria may be decreased by demethylation reactions and volatilization of dimethylmercury [15].

Health Effects

The problem with methyl-mercury is that it is consumed by aquatic organisms, especially fish and bioaccumulates in their tissues. Biomagnification of methyl-mercury poses a serious human health risk which was first realized during the 1950 and 1960's at Minamata Bay, Japan where more than 1000 people were killed and 5000-6000 suffered irreparable neurological damage from the consumption of mercury contaminated seafood. Contamination at Minamata Bay resulted from organic mercury runoff produced by an acetaldehyde facility [22].

Mercury poses such a huge threat to human health because once it enters the body the destruction that occurs is usually irreversible. Symptoms associated with mercury toxicity are

tremors, ataxia, paresthesia, sensory disturbances, cardiovascular collapse, severe gastrointestinal damage, irreversible damage to the brain, kidneys, and developing fetuses, and even death [22].

Studies conducted have shown that neurological symptoms caused by methyl-mercury can continue indefinitely even after exposure from the source has ceased [15].

3.0 Overview of Phytoremediation

Due to the extreme consequences, environmental contamination with heavy metals, particularly lead and mercury, is a significant concern. Now faced with these overly extensive environmental problems, a cost-effective means of remediation pertinent to the contaminated areas must be found. There are a number of conventional remediation technologies which are employed to remediate environmental contamination with heavy metals such as solidification, soil washing and permeable barriers. But a majority of these technologies are costly to implement and cause further disturbance to the already damaged environment. Phytoremediation is evolving as a cost-effective alternative to high-energy, high-cost conventional methods. It is

considered to be a "Green Revolution" in the field of innovative cleanup technologies.

3.1 What is Phytoremediation?

Phytoremediation is the use of green plants to clean-up contaminated hazardous waste sites. The idea of using metal-accumulating plants to remove heavy metals and other compounds was first introduced in 1983, but the concept has actually been implemented for the

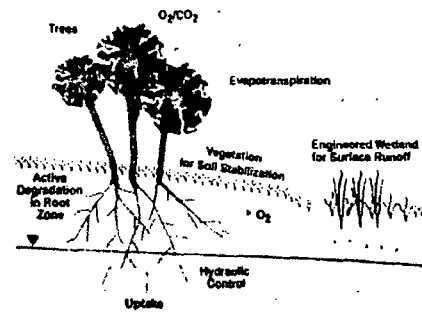


Fig. 2: Plant Processes Leading to Environmental Remediation

Source: Phytoremediation: A Growing Field with Some Concerns
http://www.the-scientist.com/yr1999/mar/black_pl_990301.html

past 300 years on wastewater discharges [5]. A general, visual reference concerning plant-based mechanisms used to remediate the environment is shown in Figure 2.

Phytoremediation has the potential to clean an estimated 30,000 contaminated waste sites throughout the US according to the EPA's Comprehensive Environmental Response Compensation Liability Information System (CERCLIS) [20]. Sites included in this estimate are those that have either been owned or contaminated by: battery manufacturers; electroplating, metal-finishing; and mining companies. Also included in the estimate are producers of solvents, coated glass, paints, leather, and chemicals [20]. Phytoremediation is aimed at providing an innovative, economical, and environmentally-friendly approach to removing toxic metals from hazardous waste sites [22].

The foundation of phytoremediation is built upon the microbial community, and the contaminated soil/water environment [25]. Complex biological, physical, and chemical interactions that occur within the soil allow for the remediation of contaminated sites. Of major importance is the interaction that takes place in the soil adjacent to the roots, called the rhizosphere. It has been shown that the rhizosphere contains 10-100 times the number of microorganisms per gram than unvegetated soil. Plants exude from their roots a variety of organic compounds that support the microbial community and facilitate the uptake of some metals [25]. The complex interactions among the roots, microbes, metals, and soil make phytoremediation a highly site-specific technology. The agronomic principles of each site must also be reviewed in order to accomplish an effective application of the technology [27].

3.2 Advantages of Phytoremediation

A significant advantage of phytoremediation is that a variety of organic and inorganic compounds are amenable to the phytoremediation process (see Table 1). Phytoremediation can be used either as an *in situ* or *ex situ* application [22, 27]. *In situ* applications are frequently

considered because minimizes disturbance of the soil and surrounding environment and reduce the spread of contamination via air and waterborne wastes. Another advantage of phytoremediation is that it is a green technology and when properly implemented is both environmentally friendly and aesthetically pleasing to the public [22].

Phytoremediation does not require expensive equipment or highly-specialized personnel, and it is relatively easy to implement. It is capable of permanently treating a wide range of contaminants in a wide range of environments. However, the greatest advantage of phytoremediation is its low cost compared to conventional clean-up technologies [27, 22]. For example, the cost of cleaning up one acre of sandy loam soil with a contamination depth of 50 cm with plants was estimated at \$60,000-\$100,000 compared to \$400,000 for the conventional excavation and disposal method [23].

Table 1: Substances Amenable to the Phytoremediation Process

Organics	Inorganics
Chlorinated Solvents TCE, PCE, MTBE, carbon, tetrachloride	Metals B, Cd, Co, Cr, Cu, Hg, Ni, Pb, Zn
Explosives TNT, DNT, RDX, and other nitroaromatics	Radionuclides Cs, ³ H, Sr, U
Pesticides atrazine, bentazon, and other chlorinated and nitroaromatic chemicals	Others As, Na, NO ₃ , NH ₄ , PO ₄ , perchlorate (ClO ₄)
Wood Preserving Chemicals PCP and other PAH's	

Source: D. Glass Associates

3.3 Disadvantages and Limitations of Phytoremediation

In contrast to its many positive aspects, phytoremediation does have a few disadvantages and limitations. It is restricted to the rooting depth of remediative plants. Remediation with plants is a lengthy process, thus it may take several years or longer to clean up a hazardous waste site, and the contamination may still not be fully remediated [27]. The use of invasive, nonnative species can affect biodiversity. The consumption of contaminated plants by wildlife is also of concern. Harvested plant biomass produced from the process of phytoextraction may be classified as a RCRA hazardous waste; therefore subject to proper handling and disposal. Unfavorable climate is another important consideration because it can limit plant growth and phytomass production, thus decreasing process efficiency [28].

Table 2: Advantages and Disadvantages/Limitations of the Phytoremediation Process

Advantages	Disadvantages / Limitations
Amendable to a variety of organic and inorganic compounds	Restricted to sites with shallow contamination within rooting zone of remediative plants
<i>In Situ / Ex Situ</i> Application	May take up to several years to remediate a contaminated site
<i>In Situ</i> applications decrease the amount of soil disturbance compared to conventional methods	Restricted to sites with low contaminant concentrations
Reduces the amount of waste to be landfilled (up to 95%)	Harvested plant biomass from phytoextraction may be classified as a RCRA hazardous waste
<i>In Situ</i> applications decrease spread of contaminant via air and water	Climatic conditions
Does not require expensive equipment or highly specialized personnel	Introduction of nonnative species may affect biodiversity

Advantages	Disadvantages / Limitations
Easy to implement and maintain	Consumption of contaminated plant tissue is also of concern
Low-cost compared to conventional treatment methods	
Environmentally friendly and aesthetically pleasing to the public	

3.4 Current Status and Projected Market of Phytoremediation

The science of phytoremediation has shown promising results as an innovative cleanup technology. However, it is still in a developmental stage and more research is needed to increase the understanding and knowledge of this remediation technology. One of the major problems encountered with the phytoremediation of heavy metals is their decreased bioavailability to plants. Generally, soil amendments are added to the soil to increase the bioavailability of heavy metals to enhance the uptake by plants. A number of environmental concerns pertaining to the use of soil amendments have arose and will be addressed in a later section. Ongoing bench-scale studies and field demonstrations are being conducted throughout the United States in order to better understand and implement this technology. As phytoremediation progresses it is expected to increase its share in the environmental cleanup market. D. Glass Associates, Inc. has already estimated a projected market for the field of phytoremediation. For 1998, the projected market was \$16.5-\$29.5 million, the year 2000 market was estimated at \$55-\$103 million, and by the year 2005, it has been estimated to reach \$214-\$370 million [9].

3.5 Environmental Concerns Associated with Phytoremediation

There are a number of environmental concerns pertaining to the use of phytoremediation. One of the most significant of these involves human health. Will the implementation of phytoremediation have an effect on the food chain? There are a number of different routes of exposure that must be taken into consideration. The ingestion of heavy metals through contaminated soil by humans or animals, ingestion of vegetation grown on the metal-contaminated soil, ingestion of animals that have ingested plants grown in the metal-contaminated soil, and the leaching of metals into the water supply are all concerns [7]. The question has been raised about how metal accumulating plants will impact the food chain through herbivores and insects that ingest the toxic plant biomass. For example, will the contaminants be present in the pollen after phytoremediation and then be dispersed to different regions by bees and other insects? Or will insects ingest the toxic metals introducing them into the food chain?

Some studies have found that certain animals and insects will not consume plants being used for phytoremediation because they merely taste bad [7]. Field observations of livestock in areas where naturally occurring metal hyperaccumulators have been found, have shown that cattle, sheep, and goats avoid the metal rich vegetation like *Alyssum* and *Thalspi*. The seeds of hyperaccumulators are generally small and lack nutritional and food value. Thus, large mammals and birds are highly unlikely to have a diet limited to metal contaminated vegetation because of their requirement for large habitats, which increases their variety of food consumed. Concern lies with smaller mammals (i.e., deer mice) and insects (i.e., grasshoppers) that cover smaller areas of land, increasing the possibility that they could solely survive within a phytoremediation field application [7].

Other concerns include the effects caused by site preparation activities on other nearby crops and vegetation such as pesticide drift and dust, and the introduction of potential, non-native (exogenous) plant species [28]. The use of *nonnative plant species* is of concern because of the potential risk of affecting native plant biodiversity. This problem can be solved by either only using native plant species for that specific region or sterile exogenous plants.

To address the environmental concern of the potential hazard to ecological receptors during the process of phytoremediation of soils, an ecological risk assessment was reviewed and added to this report. The risk assessment was conducted on soils contaminated with depleted uranium. A ecological risk assessment pertaining to the phytoremediation of lead was not found.

3.5.1 Ecological Risk Assessment

To address the environmental concern of the potential hazard phytoremediation poses to ecological receptors, Edenspace Systems Corporation contracted Risked-Based Remedies (RBR) Consulting, Inc. to conduct an ecological risk assessment study [16]. The study area was located in Aberdeen, MD at the Bomb Throwing Device (BTD) Area of the Aberdeen Proving Ground and was conducted over a five month period (June through Oct., 1999). The subject soils were contaminated with depleted uranium (DU).

The ecological risk assessment involved both the actual test plot and a selected reference area. The reference area was located about 50 feet away from the test plot and was covered with natural vegetation. The test plot was planted with Indian mustard (2 crops), and soil amendments were added to increase the mobility and plant uptake of DU. To assess the potential increase in hazard to the ecological receptors, samples were taken from surface soils, plants in both the test and reference areas, invertebrate herbivores (e.g., grasshoppers), invertebrate predators (e.g.,

spiders), and vertebrates were collected. Avian and mammalian vertebrates that were evaluated in this study were white-tailed deer, wild turkey, red fox, deer mouse, and American robin.

Initial surface soil samples revealed 102 mg/kg of DU in the test plot and 199 mg/kg of DU in the reference area. Indian mustard DU concentrations in the test plot were 2.32 mg/kg and 2.22 mg/kg during the normal growth phase (2 test periods), no addition of chemical chelate. During the hyperaccumulation phase, addition of chemical chelate, plants accumulated 14.4 mg/kg and 15.7 mg/kg of DU. The DU concentration ranged from 0.113 to 0.612 mg/kg for the natural vegetation found in the reference area. Invertebrate herbivore samples collected in the test plot showed DU levels ranging from 0.288 to 1.85 mg/kg and 0.431 to 9.16 mg/kg of DU were found in invertebrates in the reference area. The concentrations found in invertebrate herbivores (grasshoppers) was proportional to the DU concentrations found in the reference area, indicating that bioaccumulation and bioconcentration was not occurring. Toxicity data on terrestrial invertebrates from DU is non-existent, but based on a visual evaluation, no observable adverse effects were noted. Although visual data has shown no visible effects, without toxicity data on terrestrial invertebrates from DU a potential for an increase in hazard to upper trophic levels of the food web may still exist.

Based on a number of different site-specific factors and modeling calculations, species-specific exposure modeling was required to characterize the potential hazard to avian and mammalian wildlife that were not sampled directly, the Average Daily Dose from Ingestion (AAD_i) in mg/kg per day, Reference Toxicity Dose (RTD), and an ecological quotient which is a ratio of the estimated dose to the reference toxicity dose were estimated for the evaluation of wildlife. These values are located in Table 3.

The results of the ecological risk assessment revealed that the potential for adverse effects such as increased mortality or decreased reproduction, to wild turkey, white-tailed deer, red fox, deer mouse, and American robin due to consumption of DU enriched biomass is negligible. All site-specific factors were taken into consideration for this study, although some uncertainties are inherent. Problem formulation, use of the no-effect concentration (i.e., NOAEL), limited data for upper trophic levels, ingestion of Indian mustard by invertebrate herbivores, and the focus of the ecological risk assessment on trophic pathways are the major sources of uncertainty found for this particular ecological risk assessment [26].

Table 3: Calculations of Ecological Quotients

Wildlife	Depleted Uranium - Reference Area			Depleted Uranium - Test Plot		
	ADD _i (mg/kg-d)	RDT (mg/kg-d)	EHQ	ADD _i (mg/kg-d)	RDT (mg/kg-d)	EHQ
White-tailed Deer	0.00031	0.467	0.00067	0.0011	0.467	0.0023
Red Fox	0.00092	0.874	0.0010	0.00080	0.874	0.00091
Deer Mouse	0.087	3.35	0.26	0.080	3.35	0.24
Wild Turkey	0.000030	16.0	0.000019	0.00010	16.0	0.000063
American Robin	2.9	16.0	0.15	2.3	16.0	0.15

Source: Ecological Risk Assessment conducted by Risk-Based Remedies Consulting, Inc.

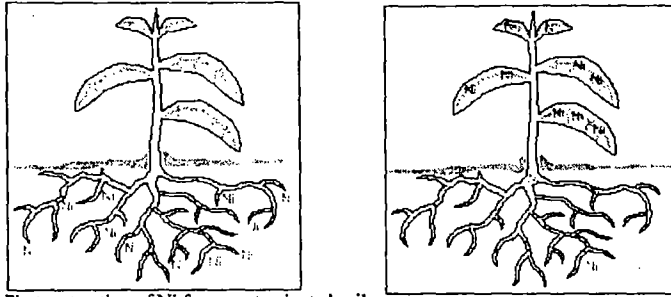
4.0 Mechanisms Used for the Phytoremediation of Heavy Metals

There are a number of different types of phytoremediation processes, which cover a large number of different organic and inorganic compounds. Only four are relevant to the phytoremediation of lead and mercury, two of the most difficult heavy metals to remove by means of phytoremediation. These four subsets of phytoremediation are termed phytoextraction, phytostabilization, rhizofiltration, and phytovolatilization.

4.1 Phytoextraction

Phytoextraction is primarily used for the treatment of contaminated soils [28]. To remove contamination from the soil, this approach uses plants to absorb, concentrate, and precipitate toxic metals from contaminated soils into the above ground biomass (shoots, leaves, etc.) (see Figure 3) [10]. Discovery of metal hyperaccumulator species demonstrates that plants have the potential to remove metals from contaminated soils. A hyperaccumulator is a plant species capable of accumulating 100 times more metal than a common non-accumulating plant. Thus, a hyperaccumulator will concentrate more than 1000 µg/g (0.1%) of Co, Cu, Cr, Pb, or 1% of Zn and Ni in their leaf dry matter [23]. Most hyperaccumulator species accumulate Ni while others have been shown to accumulate Cd, Co, Cu, Zn. Currently there are no known Pb hyperaccumulators. Certain plants can extract lead from contaminated soils, but only when certain soil amendments have been added [28]. This will be discussed in more detail in section 5.3.

There are several advantages of phytoextraction. The cost of phytoextraction is fairly inexpensive when compared to conventional methods. For example phytoremediation of a 12-acre site contaminated with lead was estimated to require 30 years and cost \$200,000 compared



Phytoextraction of Ni from contaminated soil

to \$12 million for excavation and disposal, \$6,300,000 for soil washing, and 600,000 for a soil cap [28]. Another benefit is that the contaminant is permanently removed from the soil [10]. In addition, the amount of waste material that must be disposed of is substantially decreased (up to 95% [28]) and in some cases, the contaminant can be recycled from the contaminated plant biomass [10]. The use of hyperaccumulator species is limited by slow growth, shallow root system, and small biomass production. In addition, the plant biomass must also be harvested and disposed of properly, complying with RCRA standards [10]. There are several factors limiting the extent of metal phytoextraction including:

- metal bioavailability within the rhizosphere
- rate of metal uptake by roots
- proportion of metal "fixed" within the roots
- rate of xylem loading/translocation to shoots
- cellular tolerance to toxic metals

In order for this clean-up method to be feasible, the plants must (1) extract large concentrations of heavy metals into their roots, (2) translocate the heavy metal into the surface

biomass, and (3) produce a large quantity of plant biomass [4]. In addition, remediative plants must have mechanisms to detoxify and/or tolerate high metal concentrations accumulated in their shoots [4].

4.2 Phytostabilization

Phytostabilization, also referred to as *in-place inactivation*, is primarily used for the remediation of soil, sediment, and sludges [28]. It is the use of plant roots to limit contaminant mobility and bioavailability in the soil [10]. The plants primary purposes are to (1) decrease the amount of water percolating through the soil matrix, which may result in the formation of a hazardous leachate, (2) act as a barrier to prevent direct contact with the contaminated soil and (3) prevent soil erosion and the distribution of the toxic metal to other areas [22].

Phytostabilization can occur through the sorption, precipitation, complexation, or metal valence reduction. It is useful for the treatment of lead (Pb) as well as arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu) and zinc (Zn) [28].

Some of the advantages associated with this technology are that the disposal of hazardous material/biomass is not required [28], and it is very effective when rapid immobilization is needed to preserve ground and surface waters [10]. The presence of plants also reduces soil erosion and decreases the amount of water available in the system [28]. However, this clean-up technology has several major disadvantages including: contaminant remaining in soil, application of extensive fertilization or soil amendments, mandatory monitoring is required, and the stabilization of the contaminants may be primarily due to the soil amendments [28].

Phytostabilization has been used to treat contaminated land areas affected by mining activities and Superfund sites. Three grasses have been made commercially available after a field study conducted in Liverpool, England [23]:

- *Agrostis tenuis*, cv *Parys* for copper waste
- *Agrostis tenuis*, cv *Coginan* for acid lead and zinc wastes
- *Festuca rubra*, cv *Merlin* for calcareous lead and zinc wastes

4.3 Rhizofiltration

Rhizofiltration is primarily used to remediate extracted groundwater, surface water, and wastewater with low contaminant concentrations. It is defined as the use of plants, both terrestrial and aquatic, to absorb, concentrate, and precipitate contaminants from polluted aqueous sources in their roots. Rhizofiltration can be used for Pb, Cd, Cu, Ni, Zn, and Cr, which are primarily retained within the roots [28]. An illustration of this method is shown in Fig. 4.

Sunflower, Indian mustard, tobacco, rye, spinach, and corn have been studied for their ability to remove lead from water, with sunflower having the greatest ability. In one study, after only one hour of treatment, sunflowers reduced lead concentrations significantly [22]. Indian mustard has a bioaccumulation coefficient of 563 for lead and has also proven to be effective in removing a wide concentration range of lead (4 mg/L-500 mg/L) [22, 28].

The advantages associated with rhizofiltration are the ability to use both terrestrial and aquatic plants for either *in situ* or *ex situ* applications. Another advantage is that contaminants do not have to be translocated to the shoots. Thus, species other than hyperaccumulators may be

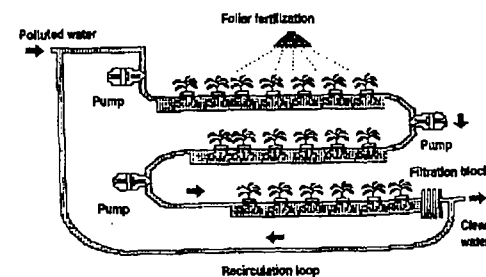


Fig. 4: Engineered rhizofiltration system
Source: Phytoremediation: Using plants to remove pollutants from the environment
<http://www.aspp.org/pubaff/phytoem.htm>

grown in a greenhouse or nursery; there is periodic harvesting and plant disposal; tank design must be well engineered; and a good understanding of the chemical speciation/interactions is needed. The cost of remediation by rhizofiltration has been estimated to be \$2-\$6 per 1000 gallons of water [28].

4.4 Phytovolatilization

Phytovolatilization involves the use of plants to take up contaminants from the soil, transforming them into volatile forms and transpiring them into the atmosphere [28]. Mercuric mercury is the primary metal contaminant that this process has been used for. The advantage of this method is that the contaminant, mercuric ion, may be transformed into a less toxic substance (i.e., elemental Hg). The disadvantage to this is that the mercury released into the atmosphere is likely to be recycled by precipitation and then redeposited back into lakes and oceans, repeating the production of methyl-mercury by anaerobic bacteria [28].

used. Terrestrial plants are preferred because they have a fibrous and much longer root system, increasing the amount of root area [22]. Disadvantages and limitations include the constant need to adjust pH, plants may first need to be

5.0 Overview of the Phytoremediation of Lead

5.1 Lead and the Soil Matrix

Once introduced into the soil matrix, lead is very difficult to remove. The transition metal resides within the upper 6-8 inches of soil where it is strongly bound through the processes of adsorption, ion exchange, precipitation, and complexation with sorbed organic matter [13, 22]. Lead found within the soil can be classified into six general categories: ionic lead dissolved in soil water, exchangeable, carbonate, oxyhydroxide, organic or the precipitated fraction. All of these categories combined make up the total soil lead content [22]. Water soluble and exchangeable lead are the only fractions readily available for uptake by plants. Oxyhydroxides, organic, carbonate, and precipitated forms of lead are the most strongly bound to the soil [6].

All of the interactions that occur throughout the soil matrix are pH dependent. The soil pH has a significant effect on the mobility of lead and other metals within the soil. The pH of soil generally ranges between 4.0-8.5. Under acidic conditions ($\text{pH} < 5.5$), metal cations are more mobile, while anions tend to sorb to mineral surfaces [13]. Metals are more available to plant roots under these conditions; however, due to an increase in aluminum (Al) solubility, plant growth may be inhibited due to Al toxicity [22]. The opposite occurs when basic conditions are present within the soil matrix. Anions are mobilized and cations are adsorbed to mineral surfaces or precipitate, decreasing the metal bioavailability for plant uptake [13]. The capacity of the soil to adsorb lead increases with increasing pH, cation exchange capacity (CEC), organic carbon content, soil/water Eh (redox potential) and phosphate levels [31].

5.2 Phytoextraction of Lead

In the natural setting, lead hyperaccumulation has not been documented. However, certain plants have been identified which have the potential to uptake lead. Many of these plants belong to the following families: Brassicaceae, Euphorbiaceae, Asteraceae, Lamiaceae, and Scrophulariaceae. *Brassica juncea*, commonly called Indian Mustard, has been found to have a good ability to transport lead from the roots to the shoots, which is an important characteristic for the phytoextraction of lead [28].

The phytoextraction coefficient for Indian Mustard (*Brassica juncea*) is 1.7 and it has been found that a lead concentration of 500 mg/L is not phytotoxic to this Brassica species [28]. A phytoextraction coefficient is the ratio of the metal concentration found within the surface biomass of the plant over the metal concentration found in the soil. Thus, the greater the coefficient, the greater the uptake of contaminant [27]. Some calculations indicate that *Brassica juncea* is capable of removing 1,550 kg of lead per acre [8].

Thlaspi rotundifolium ssp. *Cepaeifolium*, a non-crop Brassica, commonly known as Pennycress, has been found to grow in soils contaminated with lead (0.82%) and zinc from a mine. Bench scale studies have also shown that certain crop plants are capable of phytoextraction. Corn, alfalfa, and sorghum were found to be effective due to their fast growth rate and large amount of biomass produced [28].

5.3 Role of Synthetic Chelates in Phytoremediation

One major factor limiting the potential for lead phytoextraction is low metal bioavailability for plant uptake [22]. To overcome this limitation, synthetic chemical chelators may need to be added to the contaminated soil to increase the amount of lead that is bioavailable

for the plants. The use of synthetic chelates in the phytoremediation process is not only to increase heavy metal uptake by plants through increasing the bioavailability of the metal, but also to increase micronutrient availability, which decreases the possibility of plant nutrient deficiencies [2]. The goal of commercial phytoextraction is to remove or reduce the level of toxic

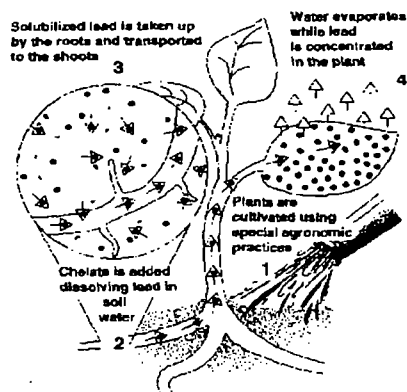


Fig. 5: Enhancement of plant uptake of Pb through the use of synthetic chelates
 Source: Phytoremediation: Using plants to remove pollutants from the environment
[Http://www.aspp.org/pubaff/phytoem.htm](http://www.aspp.org/pubaff/phytoem.htm)

metals within the contaminated soils to meet regulatory standards within 1 to 3 years [22]. The regulatory standards for lead contaminated soils set by the EPA is ≤ 500 ppm [31]. Plants that accumulate more than 1% of the target contaminant in the harvestable portion and produce more than 20 metric tons of shoot biomass per hectare per year are required to achieve this goal [22].

Researchers have found that through the application of soil amendments and chemical chelates this goal can be achieved. Based on scientific studies, it has been shown that only 0.1% of the total amount of lead in contaminated soils is in solution and bioavailable to plants for remediation. With the addition of synthetic chelators, the total amount of lead in solution can be increased up to 100 times[22].

Increasing the mobility and bioavailability of lead in the soil through certain chelators, organic acids, or chemical compounds, allows for the hyperaccumulation of metals in some plants. For lead, a number of different chelators have been tested: EDTA (ethylene-dinitri-

lo-tetraacetic acid), CDTA (trans-1,2-cyclohexylene-dinitriolo-tetraacetic acid), DTPA (diethylenetrinitriolo-pentaacetic acid), EGTA (ethylebis(oxyethylenetrinitriolo)-tetraacetic acid), HEDTA (hydroxyethyl-ethylene-dinitriolo-triacetic acid), citric acid, and malic acid [27]. Addition of the chelates resulted in enhanced shoot lead concentrations. EDTA proved to be the best and least expensive, costing around \$1.95 per pound [2, 7]. In soils with a pH of 5 and amended with EDTA, plants accumulated nearly 2000 mg/kg more lead in their shoots when compared to other treatments in soil limed to a pH of 7.5. EDTA, DTPA, and CDTA all achieved shoot lead concentrations of more than 10,000 mg/kg [2].

In order for substantial lead accumulation ($> 5,000$ mg/kg) to occur in the shoots, the concentration of synthetic chelates (EDTA, DTPA, CDTA) exceeded 1 mmol/kg. It was also noted that plants grown in soils amended with chelators varied in their lead concentration uptake. For example, the lead concentration in peas (*Pisum sativum* L. cv Sparkle) was 11,000 mg/kg compared to corn, which accumulated 3,500 mg/kg in soils receiving equivalent amounts of EDTA [2]. Although there are some advantages associated with the use of synthetic chelates, environmental concerns governing their impact on these contaminated sites are in need of research. The major concern associated with using chelates to enhance phytoremediation and increase the bioavailability of the toxic metals is the fear of lead leaching or running off into the ground or surface water. By making the metals more soluble in the soil matrix, leaching is more probable, threatening the contamination of nearby water sources [24].

5.4 Green House Study of Phytoextraction Using Synthetic Chelates

Green-house studies were conducted on soil samples collected from the Sunflower Army Ammunition Plant (SFAAP), located in the Northwest corner of Johnson County, Kansas in

order to evaluate the effectiveness of phytoextraction of ionic lead [30]. The area had been used for the production of propellants, smokeless powder, and ammunitions. Ammunition firing ranges and explosive disposal sites are also located on the SFAAP.

Two soil sampling sites, Cell 1 and 7, were involved in the green-house study. Soil lead levels ranged from 1,720 mg/kg to 3,200 mg/kg in the soil from Cell 1 and from 362 mg/kg to 3,660 mg/kg in the soil from Cell 7. The majority of lead was in the ionic form, mostly consisting of the carbonate and cerussite fractions, which are present as insoluble salts or solid phase compounds. The soil from Cell 1 was composed of an alluvial silty clay (50/50%) with a pH of 7.0. The soil samples collected from Cell 7 consisted of an alluvial silt loam (60% silt/ 25% sand/ 15% clay) with a pH of 7.3.

EDTA, EGTA, and CDTA were the three synthetic chelates tested in this experiment to enhance the mobilization and plant uptake of lead by the selected crops. It was found that EDTA was the most effective and solubilized an average of 60% of the total soil lead when applied at a rate of 15 mmol/kg at both a natural pH and under acidic conditions (pH=5.5). Both warm-season and cool-season crops were selected for the green-house study. Corn (*Zea mays* L.), sunflower (*Helianthus annuus* L.), and sorghum sudan grass (*Sorghum sudanense* L.) were studied as warm season crops and White mustard (*Brassica hirta* L.), Indian mustard (*Brassica juncea* L.), and alfalfa (*Medicago sativa* L.) were the cool season candidates.

Results gained from the green-house study concluded that of the warm-season crops, corn and sunflower were the most efficient. On average, 85% of lead concentrations were found in the harvested corn. For the cold season crops, alfalfa accumulated the highest lead concentrations in the shoot tissues at a pH of 5.5 for soils from Cell 1. These results were not representative for the soils from Cell 7. White mustard was the most effective at extracting lead

from the soils collected from Cell 7 at an EDTA-to-lead ratio of 1.5 and soil pH adjustment (these results were non-applicable to the soils collected from Cell 1). Lead concentrations found in White mustard and Indian mustard were 1.5 % by weight, a generally higher concentration than that found by other investigators. In addition, very little leachate was collected from the soil columns and only a small amount was found within the plant tissues. Thus, it is believed that most of the EDTA was bound within the soil matrix [30].

6.0 Summary of Recent Field Applications Involving Lead

To gain a better understanding concerning the current status of the phytoremediation of lead, this report provides summaries of recent field applications that have been conducted by government agencies and private phytoremediation companies. A quick overview of the field applications are available in Table 4.

6.1 Bayonne, New Jersey

An industrial site located in Bayonne, New Jersey was contaminated by cable manufacturing operations, resulting in high levels of total lead in the soil [2]. Before the implementation of phytoremediation, the lead concentration in the surface soil (0-15 cm) ranged from 1,000 to 6,500 mg/kg, with an average of 2,055 mg/kg. In the subsurface soils, at a depth of 15-30 cm, lead concentrations were lower, ranging from 780 to 2,100 mg/kg, with an average of 1,280 mg/kg and concentrations ranged from 280 to 8,800 mg/kg at a depth of 30-45 cm.

To address this problem, in 1996 a field demonstration was conducted by Edenspace Systems Corporation (formerly known as Phytotech, Inc.) to investigate the use of phytoremediation as an innovative technology. In an attempt to quantify lead leaching and to achieve a mass balance the top six inches were excavated and placed in a 3.5 feet deep lysimeter. The soil was alkaline (pH 7.9) and consisted of a sandy loam containing 2.5% organic matter. A significant fraction (66%) of the soil lead was present as carbonate.

To enhance the mobility and bioavailability of lead in the soil, amendments containing EDTA were applied at a rate of 2 mmol/kg through an irrigation system. *Brassica juncea* was the plant of choice for this particular site. Three crops were grown and each one was harvested after six weeks of growth. By the end of the growing season, the three crops of *B. juncea* had

reduced the soil lead concentration from 2,300 to 420 mg/kg in the surface soil, averaging 960 mg/kg. In the subsurface soils at the 15-30 cm depth, the average concentration of lead had been decreased to 992 mg/kg from the initial 1,280 mg/kg, but there was relatively no change at the 30-45 cm depth.

The average soil lead reduction in the surface soil from 2,055 to 960 mg/kg was substantial. It was unexpected that three crops in one growing season could remove such high amounts of lead. None of the areas at the Bayonne, New Jersey site were fully remediated below regulatory limits within the first year, but the results demonstrate the potential of phytoremediation to reduce lead levels in contaminated soils. Leaching of neither lead nor EDTA was observed as a result of the addition of EDTA to the soil. This indicates that the reduction in soil lead concentrations was due to removal by plants and not by leaching through the soil profile [2].

Table 4: Summary of Recent Field Applications Involving Lead

Site	Contractor/ Vendor	Type of Application	Initial Contaminant Concentrations	Performance/ Contaminant Removal	Site Conditions	Plant Species/ Number of Crops
Bayonne, New Jersey	Edenspace Systems Corporation	Phytoextraction with EDTA	Surface Soil (0-15 cm): 1,000 to 6,500 mg/kg Average: 2,055 mg/kg Subsurface Soil (15- 30 cm): 780-2,100 mg/kg Average: 1,280 mg/kg	- soil lead levels were reduced in surface soils from 2,300 to 420 mg/kg - soil lead levels were reduced in subsurface soils from 1,280 to 992 mg/kg	- soil was alkaline (pH=7.9) - soil consisted of a sandy loam	- <i>Brassica juncea</i> (Indian mustard) - 3 crops were grown and harvested
Dorchester, Maine	Edenspace Systems Corporation	Phytoextraction with EDTA	Surface Soil (0-15 cm): 640 to 1,900 mg/kg Average: 984 mg/kg Subsurface Soil (15- 30 cm): Average: 538 mg/kg	- total soil lead concentrations were reduced from an average of 984 mg/kg to 644 mg/kg in the surface soil - lead levels increased slightly from 538 to 671 mg/kg in the subsurface soil	- soil was acidic (pH ranged from 5.1 to 5.9) - soil consisted of a sandy loam	- <i>Brassica juncea</i> (Indian mustard) - 3 crops were grown and harvested
Trenton, New Jersey	Edenspace Systems Corporation	Phytoextraction with EDTA	lead contamination ranged from 200 to 1,800 mg/kg	- 13% of the total soil lead in the surface was reduced from 429 to 373 mg/kg - soils that exceeded 600 mg/kg of lead were reduced to 539 mg/kg, a 21% difference	soil pH ranged from 5.1 to 7.1	- <i>Brassica juncea</i> (Indian mustard) - 3 crops were grown and harvested

-31-

Site	Contractor/ Vendor	Type of Application	Initial Contaminant Concentrations	Performance/ Contaminant Removal	Site Conditions	Plant Species/ Number of Crops
Twin Cities Army Ammunition Plant (TCAAP); Site C and Site 129-3	US Army Environment al Center	Phytoextraction with EDTA and acetic acid	Site C: averaged 2,610 ppm in the surface soil Site 129-3: averaged 358 ppm in the surface soil	- results were not as good as expected - corn only averaged lead concentrations of 0.65% and 0.13% (dry weight) - White mustard was very low, averaging 0.083% and 0.034% (dry weight) of lead	- soil had a high sand content - average annual temperature was 49.6° F	- <i>Zea mays</i> (corn) first crop - <i>Brassica</i> (White mustard) second crop
Open Burn/ Open Detonation Area at the Ensign- Bickford Company	Edenspace Systems Corporation	Phytoextraction Phytostabilizati on	Area 1: 500-5,000 mg/kg Area 2: 125-1,250 mg/kg Area 3: 500-2,000 mg/kg Area 4: 750-1,000 mg/kg Area 5: 6.5-7.5 mg/kg	- total soil lead concentrations decreased from 635 to 478 mg/kg - average plant uptake was 1000 mg/kg	- soil consisted of a silt loam - pH ranged from 6.5 to 7.5	- <i>Brassica juncea</i> (Indian mustard) First Crop - <i>Helianthus annus</i> (Sunflower) Second Crop

-32-

Site	Contractor/ Vendor	Type of Application	Initial Contaminant Concentrations	Performance/ Contaminant Removal	Site Conditions	Plant Species/ Number of Crops
Confidential Superfund Site	Not Reported	Phytoextraction	- total soil lead concentrations ave. 55,480 mg/kg, with a max. value of 140,500 mg/kg	- growth chambers were used to assess some of the plant species abilities to uptake lead - <i>Taraxacum officinale</i> extracted 1059 mg/kg of lead for the first crop and 921 mg/kg for the second crop - <i>Ambrosia artemisiifolia</i> (ragweed) extracted 965 mg/kg of lead for the first crop and 1,232 mg/kg for the second crop	- soil was alkaline (pH ranged from 7.5 to 8.1)	- <i>Agrostemma githago</i> - <i>Plantago rugelii</i> - <i>Alliaria officinalis</i> - <i>Taraxacum officinale</i> - <i>Ambrosia artemisiifolia</i> (ragweed) - <i>Acer rubrum</i> (red maple)

-33-

Site	Contractor/ Vendor	Type of Application	Initial Contaminant Concentrations	Performance/ Contaminant Removal	Site Conditions	Plant Species/ Number of Crops
Confidential Dump Site for Lead Acid Batteries	Not Reported	Phytoextraction	- total soil lead concentrations averaged 29,400 mg/kg, with a max. value of 112,500 mg/kg	lead concentrations of 1695 mg/kg were found in <i>Ambrosia artemisiifolia</i> (ragweed)	- ground cover of more than 85%	Secondary Growth: - <i>Acer rubrum</i> (red maple) - <i>Rosa multiflora</i> (multiflora rose) - <i>Ambrosia artemisiifolia</i> (ragweed) - <i>T. officinale</i> (dandelion) - <i>Alliaria officinalis</i> (garlic mustard) - <i>Plantago rugelii</i> (plantain) - <i>Acer negundo</i> L. (boxelder)

-34-

6.2 Dorchester, Maine

This contaminated site, located within a heavily populated, urban residential area, is believed to have been contaminated from paint and aerial deposition sources [2]. The total soil lead concentration found in the surface soil (0-15 cm) ranged from 640 to 1,900 mg/kg, with an average concentration of 984 mg/kg. In the subsurface soils, an average of 538 mg/kg was found at a depth of 15-30 cm, and from 30-45 cm in depth 371 mg/kg was the average soil lead concentration found.

In 1996 a phytoremediation field trial was implemented at the site by Edenspace Systems Corporation on a 1,081 sq. ft. area of land to reduce the total lead levels found within the soil. The soil was acidic, ranging from a pH of 5.1 to 5.9, and consisted of a sandy loam composed of 9% organic matter. The lead was fairly evenly distributed between all soil fractions, with the organic fraction being the highest (24%).

To increase the mobility and bioavailability of the lead within the soil matrix, EDTA was applied at a rate of 2 mmol/kg through an irrigation system. Three crops of *Brassica juncea* were grown and each one was harvested after a period of six weeks from April through October of 1996. After the completion of the field trial, phytoremediation was successful in reducing the total soil lead level from an average of 984 mg/kg in the surface soil (0-15 cm) to 644 mg/kg. In the subsurface soils, at a depth of 15-30 cm, lead levels slightly increased from 538 mg/kg to 671 mg/kg, possibly due to leaching from surface soils, and from a depth of 30-45 cm, there was a slight reduction from 371 mg/kg to 339 mg/kg.

The success in the reduction of total soil lead at this site demonstrates the potential and ability of phytoremediation [2].

6.3 Trenton, New Jersey (Magic Marker Site)

The Magic Marker site, located in Trenton, New Jersey, is listed as a Brownfield site that has been contaminated from the manufacturing of lead acid batteries and other industrial activities [1]. This facility has been abandoned since 1989. The concentration of lead contamination ranged from 200 to 1,800 mg/kg and a majority of the soil lead was considered available for plant uptake. Forty percent of the lead contaminated soil exceeded 400 mg/kg and seven percent exceeded 1000 mg/kg. In order to reduce the total soil lead levels, phytoremediation was implemented on a 4,500 sq. ft. area of land by Edenspace Systems Corporation.

The soil pH varied significantly from 5.1 to 7.1 and only 28% of the total soil lead occurred in the nonavailable residual form. EDTA was applied at a rate of 200 mmol/m² to enhance the mobility and bioavailability of lead to the plants. *Brassica juncea* was used to remediate the area. Three crops were grown during the 1996 growing season and each one was harvested after 6 weeks of growth. After the third crop of *B. juncea* was harvested, the areas that exceeded 400 mg/kg were reduced from 179m² to 128m², resulting in only 28% of the treated area exceeding 400 mg/kg compared to the initial 40%. None of the treated area exceeded 800 mg/kg and only 25m² exceeded 600 mg/kg by the end of the growing season.

The results of this study also indicated that the most significant impact of phytoremediation was on the surface soil (0-15 cm). The average lead concentration of the surface soil decreased from 429 to 373 mg/kg, resulting in a 13% reduction, even though a majority of the surface soils were initially below 400 mg/kg. In areas exceeding 600 mg/kg,

phytoremediation had a substantial impact by decreasing lead concentration from 736 to 539 mg/kg, a 21% decrease.

In an attempt to determine the possibility of downward movement of lead into the soil profile and groundwater, one section was treated with 2 mol EDTA/m² in the fall of 1996. In the spring of 1997, soil samples were taken to a depth of 120 cm to evaluate the effects of the precipitation from the previous winter on the distribution of both lead and EDTA within the soil matrix. The results revealed that the total soil lead concentration decreased with depth, 878 mg/kg was found at the surface compared to 15 mg/kg at 120 cm in depth. Soil samples also concluded that EDTA resides primarily in the upper 30 cm of the soil. It should be noted that the data was variable, suggesting a further need for research pertaining to this topic [1].

6.4 Twin Cities Army Ammunition Plant (TCAAP); Site C and Site 129-3

Twin Cities Army Ammunition Plant (TCAAP), located in Minneapolis-St. Paul Minnesota, is a 2,370 acre facility that has been involved in the production of small arms ammunition, related materials, fuses and artillery shell materials [19]. In 1981, it was recognized that contaminated groundwater from TCAAP was migrating into the Minneapolis-St. Paul metropolitan groundwater supply. The total soil lead concentration ranged from an average of 2,610 ppm in the surface soil at site C to an average of 358 ppm in the surface soil of site 129-3.

The soil at site C was peat, underlain by fine sand and sandy clay. Site 129-3 was composed of fine to medium grained sand. Both sites contained high volumes of sand, creating an opportunity to observe potential leaching. Due to the average annual temperature of 49.6° F at this site, the climate was less than ideal for growing crops. This provided an excellent

opportunity to examine the operational feasibility of phytoremediation in non-optimal climatic conditions.

Corn was the first crop grown on both sites and white mustard was the second. Soil amendments containing EDTA and acetic acid were applied to the soil to enhance the mobilization and uptake of lead. The results obtained from this field demonstration were not as promising as expected. The first crop, corn, yielded only 2.1 to 3.6 tons of biomass per acre compared to the anticipated yield of 6 tons per acre, and the dry weight lead concentrations averaged 0.65% and 0.13% for sites C and 129-3, respectively. White mustard, the second crop, yielded 1.9 to 2.1 tons of biomass per acre where it was capable of growing. Average dry weight lead concentrations were low, 0.083% and 0.034% for sites C and 129-3, respectively.

A preliminary cost estimate was provided for this site which estimated \$30.34 per cubic yard of soil per year or \$153 per cubic yard of soil for the entirety of the project [19]. For further information, a complete case study can be found online at <http://bigisland.tclients.com/frtr/00000175.html>.

6.5 Open Burn/Open Detonation Area at the Ensign-Bickford Company

The OB/OD area at the Ensign-Bickford Company, located in Simsbury, Connecticut located within the 100 year flood plain of the Farmington River, has been highly contaminated with lead due to past activities [18]. From 1996-1997, a full-scale phytoremediation project was demonstrated by Edenspace Systems Corporation on 1.5 acres surrounding the open burn, open detonation area. Successful results were obtained for 1997, which resulted in the increasing of the project to 2.35 acres in 1998, combining both phytoextraction and phytostabilization.

The 2.35 acres was divided into five treatment areas. The total soil lead concentrations were as follows: Area 1 ranged from 500-5,000 mg/kg, area 2: 125-1,250 mg/kg, area 3: 500-2,000 mg/kg, area 4: 750-1,000 mg/kg, and area 5 ranged from 6.5-7.5 mg/kg. Areas 1 through 4 were treated using phytoextraction and phytostabilization was implemented at area 5. The soil was composed of a silt loam and ranged in pH from 6.5 to 7.5. Soil amendments were applied to areas 1 through 4 to increase the mobility of the lead within the soil profile. Three crops were planted and harvested for the 1998 growing season. The first crop grown was Indian mustard (*Brassica juncea*), the second was sunflower (*Helianthus annuus*), with the third crop a combination of both Indian mustard and sunflower. Plant growth was considered generally good for the 1998 crops, especially taking into consideration the high water table. However, certain areas remained excessively wet, resulting in poor plant growth and decreased biomass production.

Phytoextraction in areas 1 through 4 resulted in a decrease in total soil lead concentrations from an initial average of 635 mg/kg (April 1998) to 478 mg/kg (October 1998). After the 1998 growing season, no soil samples taken exceeded 4000 mg/kg. Before phytoextraction had been implemented, 7% of the treatment area had soil lead concentrations in excess of 2000 mg/kg and after the treatment process only 2% still exceeded that amount. The lead uptake in Indian mustard ranged from 342 mg/kg (weight) for the first crop to 3,252 mg/kg for the third crop. The average lead uptake was similar in both sunflower and Indian mustard with a value of 1000 mg/kg in the sunflower and 1,091 mg/kg (dry weight) in Indian mustard [18]. For more information regarding this site, please contact Dr. Michael Blaylock of Edenspace Systems Corporation by phone at (703)390-1100 or Email: SoilRx@aol.com. For

information regarding the 1998 field study a complete case study can be located online at <http://bigisland.tlclients.com/fttr/00000164.html>.

6.6 Confidential Sites

6.6.1 Superfund Site

A phytoremediation field study was conducted on a Superfund site, approximately 7.5 acres in area, contaminated over a 90 year time frame from battery recycling, foundry, and secondary smelting operations leading to significant soil contamination by lead [21]. Homes have been built immediately adjacent to the site. Total soil lead concentrations averaged 55,480 mg/kg, with maximum values reaching 140,500 mg/kg. The soil lead primarily occurred in the carbonate fraction (41.6%), with 28.9% in the sulfide/residual fraction, and 26.7% in the organic chemical fraction. It was estimated that 71.4% of the total soil lead was in the non-residual form, making it bioavailable for plant uptake. The soil at this Superfund site was alkaline with a pH range of 7.5 to 8.1.

Eighty-five percent of the area was vegetated by native plant species, predominantly grasses, legumes and assorted perennials. Due to extreme levels of total soil lead, roots of all the plants were severely stunted, generally not penetrating past a depth of 5 cm. Plants used in this study were *Agrostemma githago*, plantain (*Plantago rugelii*), garlic mustard (*Alliaria officinalis*), dandelion (*Taraxacum officinale*), ragweed (*Ambrosia artemisiifolia*), and red maple (*Acer rubrum*). Dandelion, ragweed and red maple were studied in growth chambers to assess their ability to extract lead from lead contaminated soil.

The results of the field study revealed that lead uptake by the plants varied from non-detectable to 1,800 mg/kg taken up by *Agrostemma githago* roots. In the growth chamber study,

dandelion was successful at extracting 1059 mg/kg of lead from the contaminated soil for the first crop and 921 mg/kg for the second. The first crop of ragweed was successful at extracting 965 mg/kg of lead, with an increase to 1232 mg/kg for the second crop. Each crop was allowed a growth period of 60 days. Plants that removed lead were predominantly herbaceous species, with some of them producing a sufficient amount of biomass. Of the plants studied, more than 65% of them had a higher concentration of lead in their roots than in their shoots [21].

6.6.2 Dump Site for Lead Acid Batteries

The acid battery dump site, approximately 5 acres in area, was originally used as a limestone quarry and filled with unknown materials prior to 1941 [21]. In 1993, battery casings were discovered along with what appeared to be foundry sand. The total soil lead concentrations averaged 29,400 mg/kg, with maximum values of 112,500 mg/kg of lead. 48.5% of the lead was found in the organic fraction and 7.8% in the sulfide/residual form. It is estimated that 92.2% of the total soil lead is in the non-residual fraction and is therefore bioavailable to plants.

The site is vegetated with the native growth of Red Maple (*Acer* spp.), multiflora rose (*Rosa multiflora*), ragweed (*Ambrosia artemisiifolia*), dandelion (*T. officinalis*), garlic mustard (*Alliaria officinalis*), plantain (*Plantago rugelii*) and boxelder (*Acer Negundo* L.) producing a ground cover of more than 85%. The results obtained from this field demonstration reveal undetectable lead levels in the shoots of garlic mustard and *Lepidium campestre*, compared to 1695 mg/kg in ragweed.

The most significant finding of the phytoremediation field trials at the superfund and lead acid battery dump site was the ability of the plants to survive in such highly lead contaminated soils. It was noted that the overall lead uptake by plants on the Superfund site was greater due to

the increased amount of soluble lead in the soil. The phytoremediation potential of dandelion and ragweed was also noted based on the results obtained from the growth chamber studies for their ability to extract lead from contaminated soils [21]. These field trials are on-going and to find out more information pertaining to these sites please contact John Pichtel located at Ball State University by phone (765)285-2182 or Email: jpichtel@gw.bsu.edu.

7.0 Overview of the Phytoremediation of Mercury

7.1 Mercury and the Soil Matrix

Complex interactions that occur between the soil and mercury will determine the transformation it will undergo once introduced into the soil matrix. The most common forms of mercury found at contaminated hazardous waste sites are the mercuric form (Hg^{2+}), mercurous form (Hg_2^{2+}), elemental mercury (Hg^0), and methyl or ethyl mercury (alkylated form) [32]. The methylated form of mercury (methyl-mercury) is the most toxic species, 10,000 times more than elemental mercury [25]. Under oxidizing conditions within the soil, Hg^{2+} and Hg_2^{2+} are the most stable forms. The cations of mercuric Hg and mercurous Hg are adsorbed by negatively charged clay minerals, oxides, and organic matter which increases the pH of the soil and therefore, increasing the stabilization [32].

The mercurous form (Hg_2^{2+}) of mercury precipitates when in the presence of chloride, phosphate, carbonate, or hydroxide, resulting in stabilization and decreased mobility. In a mild reducing environment, the toxicity and mobility of mercury is increased by the transformation of organic and inorganic mercury into methyl or ethyl mercury through biotic and abiotic processes. In the presence of a strong reducing environment, HgS is the predominant mercury compound, which is also fairly insoluble and non-toxic [32].

The form of mercury that has received the most attention and concern is methyl-mercury. Methyl-mercury is produced by methylating bacteria which are most commonly found in aquatic systems and sediments. Methyl-mercury is about 50 times more toxic than ionic mercury and can be biomagnified up to 160 fold when introduced into the food chain. Some bacteria have shown to possess the ability to protect themselves against toxic methyl-mercury [14].

7.2 Phytoremediation of Mercury

7.2.1 Transgenic Plants

There is some evidence that certain plant species have the ability to extract and accumulate mercury both from the atmospheric and soil sources [22]. Currently, however, no plant species with mercury hyperaccumulating properties has been identified [22]. Due to this, scientists have been researching the use of genetically engineered plants by inserting bacterial genes specific for detoxifying toxic forms of mercury.

Mercuric ion reductase (*merA*) and organomercurial lyase (*merB*) are two bacterial genes used for detoxifying methyl-mercury. Genetic engineering has been used to transfer bacterial genes, *merA* and *merB*, into plants. Expression of *merB* catalyzes the release of Hg^{2+} from the organic compound. Subsequently, *merA* catalyzes the reduction of Hg^{2+} taken up by the plants to elemental Hg, which is volatilized in the atmosphere. Thus, transgenic plants, *Arabidopsis thaliana L.* and tobacco (*Nicotiana tabacum*) containing both the *merA* and *merB* bacterial genes have the ability to transform methyl-mercury into elemental mercury, releasing it into the atmosphere through a process termed phytovolatilization [14].

From a regulatory perspective, however, Hg released into the atmosphere is not acceptable and the use of plants genetically altered with *merA* and *merB* genes is not permitted. There has been some efforts to use only plants transformed with *merB*. The bacterial gene *merB* seems to allow for the germination and survival of plants on a medium contaminated with organomercurial compounds, phenyl mercuric acetate (PMA), or methyl-mercury [22]. The *merB* gene allows plants to transform methyl-mercury into less toxic Hg^{2+} , preventing the

introduction of methyl-mercury into the food chain as well as preventing Hg volatilization into the atmosphere [14].

A research team headed by Richard Meager at the University of Georgia has started to investigate the use of trees in the phytoremediation of mercury. In the laboratory they have successfully developed a yellow poplar (*Liriodendron tulipifera*) that is not only fast growing and pest resilient, but also effective at absorbing and releasing mercury vapor at a rate of up to 10 times that of control plants [14]. In addition to merA and merB, the bacterial genes merP and merT (mercury transport genes) need to be researched because they may have significant potential for improving mercury uptake and translocation to specific organelles and tissues [22].

7.2.2 Phytoextraction of Mercury

Hope for the phytoextraction of mercury has been inspired by recent discoveries showing some plants produce specific peptides, termed phytochelatins, that bind and detoxify hazardous metals such as cadmium [15]. Phytochelatins were discovered in *Arabidopsis* and other mustard species, after initially being screened in wheat roots. Researchers speculate that if the amount of phytochelatins within a metal accumulating plant could be increased, the level of contaminant removed by plants would also increase [15]. Substantial evidence indicates that phytochelatins play a role in Cd phytoremediation. However, more research is needed to determine whether phytochelatins also play a role in Hg²⁺ phytoremediation.

7.3 Bench Scale Study: Argonne National Laboratory West, Waste Area Group 9, Operable Unit 9-04 (ANL-W)

Argonne National Laboratory West, Waste Area Group 9, Operable Unit 9-04 (ANL-W) is part of the Idaho National Engineering and Environmental Laboratory (INEEL), a government facility managed by the Department of Energy (DOE) [17]. ANL-W was used for scientific and

engineering research, contaminating it with petroleum products, acids, bases, PCBs, radionuclides, and heavy metals. The primary contaminants are mercury, chromium, selenium, silver, and zinc.

Bench-scale studies were conducted on the contaminated soil to address the potential of phytoremediation as a remedial alternative. The total concentration of mercury within the soil was less than 1.5 mg/kg. The soil was composed of a sand (47%), silt (34.6%), and clay (18.4%) loam with a pH of 8.57. Candidate plant species selected for this study included: Prairie Cascade hybrid willow (*Salix x*), canola (*Brassica napus*), and kochia (*Kochia scoparia*).

Results obtained from the bench-scale study indicated that an optimum formulation of chelating agents would be a 0.05 molar solution of 40% EDTA and 60% citric acid. The hybrid willows had the best recovery of mercury (42%) in the sand experiments and the recovered Hg was found almost exclusively in the roots. There was no mention concerning the plant uptake of mercury from the contaminated soil samples collected from the waste site. The estimated cost for the implementation of a phytoremediation field demonstration of a 2-year project was \$300,000. To extend it for five more years the estimated cost was \$542,000 and a maximum cost of \$780,000 was estimated for a 20-year remedial project [17].

Based on the bench-scale studies, phytoremediation is a potential cleanup method that could be used at ANL-W. A two-year phytoremediation field trial is going to be implemented using three-foot tall bare-root willow trees planted in a grid pattern. At the end of each growing season the entire tree (roots and surface biomass) will be harvested, chipped and transported to an off-site incinerator for disposal.

8.0 Conclusion

Phytoremediation is the use of green plants to clean up hazardous waste sites. According to the EPA's Comprehensive Environmental Response Compensation Liability Information System (CERCLIS), phytoremediation has the potential to clean up an estimated 30,000 hazardous waste sites throughout the US. Phytoremediation is amenable to a variety of organic and inorganic compounds and may be applied either *in situ* or *ex situ*. *In situ* applications decrease soil disturbance and the possibility of contaminant from spreading via air and water, reduce the amount of waste to be land filled (up to 95%) and are low-cost compared with other treatment methods. In addition to this, it is easy to implement and maintain, does not require the use of expensive equipment or highly specialized personnel and is environmentally friendly and aesthetically pleasing to the public. However, phytoremediation is limited to sites where contamination concentration is low and near the surface, may take several years to remediate a contaminated site and the harvested plant biomass may be classified as a RCRA hazardous waste, requiring proper handling and disposal. Thus, climatic conditions are also a limitation when dealing with a biological remediation process.

There are a number of environmental concerns that are associated with the use of plants to remove toxic metals from contaminated soils. A major concern is how phytoremediation could affect the food chain through herbivores and omnivores that may ingest the metal-laden foliage. Other concerns focus on the effects caused by site preparation activities on other nearby crops and vegetation, the introduction of potential, non-native plant species and the use of synthetic chemical chelates for the phytoextraction of Pb to increase the bioavailability and plant uptake of the metal.

The use of phytoremediation to clean up lead and mercury contaminated sites has shown promising results, but is still in the research and development stage. A number of different laboratory and field studies have been conducted focusing on the phytoremediation of Pb. The results have been very promising, although further research and understanding of the process is still required. Phytoremediation studies involving mercury are still being conducted within a laboratory setting. The use of transgenic plants has sparked interest for the phytoextraction of mercury. More research is needed regarding the different bacterial genes that can be used for the phytoremediation of methyl-mercury. Bacterial genes that allow the plants to not only grow, but uptake the methyl-mercury and store it in their surface biomass are needed in order to successfully remove the toxic metal from contaminated soils.

Phytoremediation is considered to be an innovative technology and hopefully by increasing our knowledge and understanding of this intricate clean up method, it will provide as a cost-effective, environmentally friendly alternative to conventional clean up methods.

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