Improving Bioremediation of PAH Contaminated Soils by Thermal Pretreatment

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COVER

photographs from top to bottom: tar pond near oil refinery; tar contaminated soil at former cokes plant; coal tar particles in soil contaminated with creosote oil

Abstract

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Numerous sites and large volumes of sediments in the Netherlands are contaminated with polycyclic aromatic hydrocarbons (PAH), which are of great concern because of their toxic and carcinogenic effects. Since PAH tend to sorb very strongly to the soil matrix, bioremediation is a slow process with often high residual concentrations after remediation. In this study it was tried to develop methods to improve bioremediation, this means to decrease residual concentrations after bioremediation. In a literature survey, several methods were evaluated and two methods were selected for further study: soaking of soil with an organic solvent and short-term heating of soil. In biodegradation experiments with PAH contaminated soils short-term heating resulted in the largest increase of the PAH fractions that could be degraded. Desorption experiments revealed that desorption of PAH took place in two stages: a fast-desorption stage and a slow-desorption stage. The increase of the biodegradable fractions after heating in the biodegradation experiments could be related to an increase of the fraction of PAH that exhibited fast desorption. A comparison between biodegradation kinetics and desorption kinetics showed that desorption experiments could predict biodegradation results, except for high molecular weight PAH (5 or more aromatic rings). Biodegradation rates of high molecular weight PAH were always lower than desorption rates, possibly caused by slow growth of microorganisms that had to degrade these compounds. A mathematical model, describing desorption and biodegradation of PAH contaminants, showed that this slow growth could be caused by the low aqueous phase concentrations of the soil sorbed high molecular weight PAH. A technical and economical evaluation of a thermal pretreatment of PAH contaminated soils showed that a thermal pretreatment can be succesfully incorporated in current remediation practices and that it can be a competive alternative to current remediation techniques.

Table of Contents

1

General introduction

INTRODUCTION

SOIL CONTAMINATION HISTORY

Dutch soil remediation history starts in 1978 with the discovery of chemical waste in the soil under a residential area in Lekkerkerk. As a result of this affair a survey was initiated to investigate the number of other sites that had to be remediated [1]. It was estimated that approximately 350 sites were severely contaminated and should be remediated. Total costs for remediation of these sites were estimated to be more than one billion Dutch guilders. However, later investigations came up with much larger figures: in 1991 it was estimated that more than 100 thousand sites were contaminated and that total costs for remediation were 50 billion guilders [2]; a survey in 1998 gave a number of 430 thousand contaminated sites with total costs for remediation of 88 billion guilders [3]. Besides these contaminated sites of terrestic soils, large volumes of sediment have to be dredged for nautical and sanitation reasons every year. It was estimated that in the period of 1996 to 2015 880 million cubic metres of sediment have to be dredged of which 288 million cubic metres are weakly contaminated and 203 million cubic metres are severely contaminated [4]. Currently, almost no dredged sediment is remediated: weakly contaminated sediment is put on land and severely contaminated sediment is disposed in storage basins as the 'Slufter' and the 'Papegaaienbek'.

Approximately one percent of the contaminated sites contains polycyclic aromatic hydrocarbons (PAH) [3] and more than 90 percent of the sediments are contaminated with PAH [5]. PAH are a group of hydrocarbons consisting of two or more fused benzene rings. They are non-volatile, they have very low solubilities in water and they tend to sorb very strongly to the solid soil matrix. As a result, remediation of PAH contaminated soil has proofed to be very costly and energy intensive.

PAH CONTAMINATIONS

PAH contaminations of soil are of great concern since PAH can have toxic, mutagenic and carcinogenic effects. The smaller two and three ring PAH compounds show acute toxic effects on humans and animals [6,7] and chronic toxic effects on plants [6,8]. Oxidation of larger PAH compounds by human or microbial enzymes can lead to metabolites that are high;y carcinogenic [7,9].

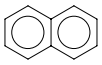
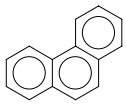
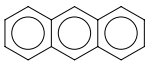
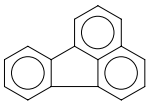
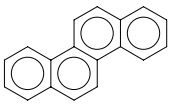
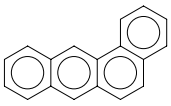
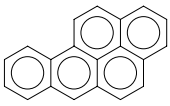
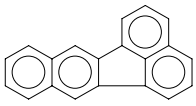
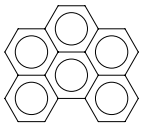
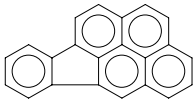
In general, PAH are formed during incomplete combustion of organic material. Through atmospheric deposition these PAH form diffuse contaminations. Natural

sources of PAH contaminations are forest fires and volcanic eruptions. Diffusive PAH contaminations from antropogenic sources are found near roads, railways, factories and within cities [7].
High concentration of PAH contaminations are found at former gas plant sites [8]. Gas plants produced coal gas by dry distillation of charcoal. This coal gas was used for heating of houses and illumination of streets. Coal gas production results in coal tar as a waste product. This coal tar consists for 30 % of PAH. Initially this coal tar was dumped in coal tar pits at the gas plant site itself. As a consequence, several hundreds of former gas plants sites in the Netherlands are severely contaminated. Later coal tar was used for the production of wood preservation agents (creosote oil) and tar coatings. Factory sites that have manufactured these products are also very often contaminated with PAH [7,10].
Also crude oil and heavy oil distillation products contain high concentrations of PAH. Leakage and dumping of oil are also sources of PAH contaminations [11].

SOIL REMEDIATION POLICY

The 'Lekkerkerk' affair in 1978 and the subsequent survey on contaminated sites in the Netherlands gave rise to the Temporarily Act on Soil Remediation in 1983 [1]. In 1995 this act has been replaced by the Remediation Regulations [12] of the Soil Protection Act [13]. Both the Temporarily Act on Soil Remediation and the Soil Protection Act stated that contaminated soil has to be restored towards multifunctionality, which means that the soil can fulfil all potential functions for both humans and ecosystems. For contaminations, multifunctionality has been defined as that contaminant concentrations are below target values (former A-values or reference values). Another level that determines the contamination situation is given by the intervention values (former C-values), above which soil is denoted as severely contaminated and remediation of the soil is required. These intervention values are determined by risks for human health and ecosystems [1,14]. Target values and intervention values are given in Table 1.1 for the ten PAH compounds that have been appointed by the Dutch Ministry of Housing and Environment (VROM).
In 1995 the Construction Materials Decision was published [15], which is related to both the Soil Protection Act and the Act for Protection of Surface Waters. This decision stated that slightly contaminated material is allowed to be used in construction works, provided that contaminant concentrations meet certain values, so-called composition levels. Composition values for PAH in soils that are used in construction works are also shown in Table 1.1.

Table 1.1 Target values, intervention values and composition values of PAH in soil

PAH compound	*chemical structure*	*target value*[a,c] *(mg/kg)*	*intervention value*[a,c] *(mg/kg)*	*composition value*[b,c] *(mg/kg)*
naphthalene		-	-	5
phenanthrene		-	-	20
anthracene		-	-	10
fluoranthene		-	-	35
chrysene		-	-	10
benzo[a]anthracene		-	-	40
benzo[a]pyrene		-	-	10
benzo[k]fluoranthene		-	-	40
benzo[ghi]perylene		-	-	40
indeno[1,2,3-cd]pyrene		-	-	40
total (sum 10 PAH)		1	40	40

[a]from Soil Protection Act [14], only for sum of concentrations of 10 PAH and not for individual compounds

[b]from Construction Materials Decision [15]

[c]values are for a standard soil with 10 weight% organic matter; these values are corrected for soils with higher organic matter contents

REMEDIATION OF PAH CONTAMINATED SOIL

REMEDIATION TECHNOLOGIES

Due to the high risk for human health and ecosystems, most PAH contaminated soils have to be remediated. Remediation of contaminated soils can be performed using either in-situ technologies or ex-situ technologies. With in-situ remediation, the contaminated soil is kept in place and is treated by injecting different compounds or agents into the soil to stimulate degradation of contaminants or by withdrawing water or air from the soil to remove contaminants. During ex-situ remediation, the contaminated soil is excavated and treated either on-site or transported and decontaminated elsewhere. For PAH contaminations, almost no in-situ remediation is currently applied because of limited perspectives and remediation results so far [3].

For ex-situ remediation, a number of technologies can currently be applied: thermal treatment, soil washing/classification, solvent extraction, and bioremediation. During thermal decontamination, soil is heated to temperatures between 400 and 800 °C, depending on the type of contaminant [16,17]. At these temperatures, organic contaminants evaporate from the soil or decompose; the evaporated contaminants are incinerated in a subsequent step.

With soil washing/classification, contaminated soil is separated in generally two fractions, based on particle size and specific weight [18,19]. The sand fraction generally contains very little soil organic matter and consequently very little hydrophobic contaminants, as these contaminants tend to sorb to organic matter. Without any further treatment, this sand fraction can mostly be re-used as construction material. The residual sludge fraction (i.e. the classification sludge) contains most of the contaminants and therefore has to be disposed of or remediated [19].

Solvent extraction removes contaminants from soil by extracting and concentrating the contaminants in an organic solvent (e.g. acetone). This technology is currently not applied in the Netherlands, although successful laboratory and pilot-scale studies have been performed [16,20,21].

Finally, in bioremediation organic contaminants are converted by micro-organisms (bacteria, fungi) to carbon dioxide and water. Bioremediation will be described in more detail in next paragraphs.

Besides remediation, large volumes of excavated contaminated soils are disposed at controlled disposal sites. The different volumes of soil that were disposed and decontaminated in 1998 using technologies described above are given in Table 1.2.

Table 1.2

Volumes of contaminated soil treated or disposed in the Netherlands in 1998

technology	*volume[a] (kton)*
thermal treatment	700
soil washing/classification	820
bioremediation	380
disposal	1250

[a] from reference [22]

BIODEGRADATION

The use of biodegradation for removal of organic contaminants from soil has two important advantages above the use of physical and chemical methods. First, the physical soil structure remains intact and the biological function of the soil is preserved in contrast to methods as thermal treatment and solvent extraction. This means that soil can be multifunctionally applied after sufficient decontamination. Secondly, bioremediation yields no residual waste products and energy consumption and operation costs are relatively low in general.

Most hydrophobic organic compounds are biodegradable, which means that they can be converted by micro-organisms to more simple compounds as carbondioxide, water and/or methane. For PAH, this degradation can be performed under aerobic [23-25] and anaerobic conditions [23,26]. Anaerobic degradation of PAH is very slow compared to aerobic degradation and, therefore, plays no significant role in bioremediation of PAH contaminated soils. PAH can be converted under aerobic conditions by both bacteria and fungi. So far, only bacteria have been used in bioremediation, although several studies towards the application of fungi, especially white-rot fungi, have been performed [27-29].

For bacterial degradation of PAH, it has been found that low molecular weight PAH compounds (with 4 aromatic rings or less) can be used as a single carbon source for growth of micro-organisms [23,24]. For high molecular weight PAH (5 aromatic rings or more), only cometabolic degradation has been found so far [23,30]. This means that these compounds are accidentally degraded by micro-organisms that use an additional substance as a primary substrate.

BIOREMEDIATION TECHNOLOGIES

Discrimination between different biological decontamination technologies can be made on basis of the energy-input during remediation (e.g. mixing, stirring, tilling) or the time required for decontamination, which is related to the mechanical energy-input. Generally, a more intensive technology leads to a

shorter treatment time. In order of decreasing intensivity and increasing treatment time, bioremediation technologies can be divided in a) soil-slurry remediation, b) biopiles and landfarming, and c) in-situ remediation.

In soil-slurry remediation, a soil-water mixture with a solids content of 10 to 20 weight percent, is intensively mixed and aerated to prevent mass transfer limitations of the contaminants, nutrients, and oxygen and to obtain maximal biodegradation rates. Treatment times for PAH contaminated soils are typically between 3 to 14 days. Longer treatment times are unacceptable for economic reasons, because of the high costs due to the large energy requirements for mixing and aeration. Soil slurry remediation can very well be applied for fast reduction of relatively high contaminant concentrations [31].

Currently, slurry remediation is not applied in the Netherlands, although several pilot scale studies have been conducted [32,33]. In the United States, there are both pilot-scale and full-scale installations for slurry remediation [34-37].

Biopiles and landfarms are more extensive treatment technologies for excavated soils, compared to slurry treatment. In both biopiles and landfarms, excavated soil is spread in layers with a thickness of 0.5 to 1.5 metres. Because the soil layer is kept aerobic, biodegradation can take place. In biopiles, the soil is regularly tilled during the remediation process and nutrients and water contents are monitored and regulated. To control temperature and precipitation, biopiles can be put in greenhouses or halls. Landfarming is a more extensive process, in which only pollutant concentrations are monitored. Landfarming can also be used as a post-treatment step for remediation in biopiles or bioreactors [31]. Typical treatment times are 6 months to 2.5 years for biopiles and up to 10 years for landfarming.

In the Netherlands, biopiles and landfarming are currently applied in full scale for remediation of terrestic soils [38,39] and in pilot scale for remediation of dredged sediments [18].

Biological in-situ remediation of contaminated soils spans a wide range of different technologies. The most important technologies will be discussed below.

Natural attenuation is the most extensive in-situ technology, without any interference regarding the remediation process. During natural attenuation, the contaminants in the soil are degraded by micro-organisms present in the soil. Only the progress of the remediation process is monitored. In case soil conditions are not favourable for natural attenuation, biodegradation can be stimulated by changing soil conditions. Possible actions are injecting air (bioventing, air sparging) [10,40] or alternate electron acceptors (e.g. nitrate, oxygen-releasing compounds) [41], adding nutrients (nitrogen, phosphate) [40,42], or increasing soil temperature (especially in arctic climates) [42,43].

In the saturated zone of the soil, the more mobile contaminants can be removed by ground water remediation, which is mostly carried out by pump-and-treat technology. Groundwater containing contaminants is withdrawn from the soil, the contaminants are (biologically) degraded, and the clean water is re-injected [41,44,45]. In the vadose zone, a similar technology can be applied for volatile contaminants by withdrawing air from the soil [44,45]. This process is called soil-venting or soil vapour extraction.
As mentioned before, degradation rates decrease from slurry remediation towards in-situ techniques. Despite of the lower degradation rates in bio-piles and landfarming compared to slurry-reactors, final decontamination efficiencies were found to be the same [46]. It can be expected that final decontamination efficiencies of in-situ bioremediation are also comparable to efficiencies of slurry remediation, biopiles and landfarms for homogeneous soils. For heterogeneous soils efficiencies will most likely be lower. It should be noted that in-situ techniques can be applied difficultly for soils with a low permeability, like clay and peat soils.

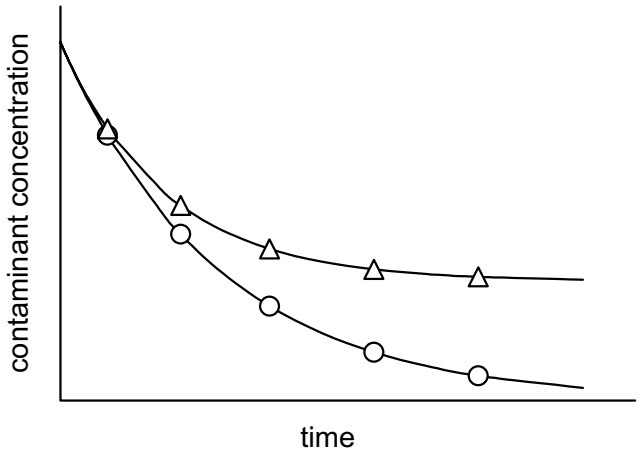
BIOAVAILABILITY

RESIDUAL CONCENTRATIONS

In the previous paragraphs it has been mentioned that bioremediation of contaminated soil has some important advantages over physical or chemical remediation technologies. However, it has very often been described that high residual contaminant concentrations remain after biodegradation, whereas physical and chemical remediation technologies remove all contaminants from the soil. Laboratory experiments showed that freshly spiked contaminants in soil could be completely degraded, while residual concentrations remained for aged contaminations [47,48]. Typical degradation curves of 'fresh' and 'aged' contaminants are shown in Figure 1.1. In some cases, these high residual concentrations might be caused by poor biodegradation, due to unfavourable environmental conditions, toxicity or absence of contaminant degrading micro-organisms. In most cases, however, residual concentrations are caused by slow desorption of the contaminants from the soil matrix [49-52].
In general, the amount of contaminants that can not be removed from soil by bioremediation depends on soil characteristics, contaminant properties and contaminant history. It has been found that residual concentrations tend to increase with increasing organic matter and clay contents of the soil [47,52,53].

Figure 1.1

Typical degradation curves of 'fresh' contaminants (○) and 'aged' contaminants (Δ) in soil



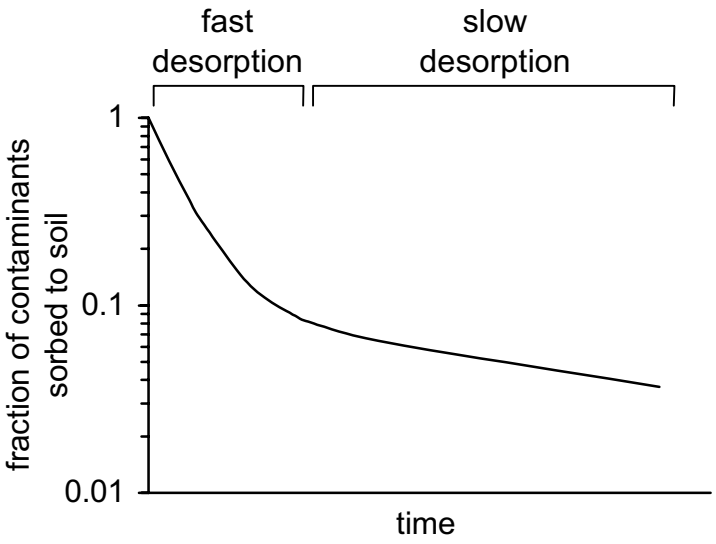
Further, residual concentrations are related to the hydrophobicity of the contaminants: the most hydrophobic contaminants show the highest residual concentrations [23,53]. Finally, increasing contact times between contaminants and soil lead to larger fractions of the contaminants that are resistive to degradation [52,54].

BIOAVAILABILITY: DEFINITION

The situation that a fraction of the contaminants is resistive to desorption or degradation is also referred to as 'limited bioavailability'. In general, the term bioavailability concerns the extend to which specific compounds are available for biota. Because this term is used in various disciplines like (eco)toxicology [55,56], agricultural sciences [57] and environmental sciences (e.g. [58-60]), the exact definition of it differs between different disciplines, but also within disciplines. Bioavailability can refer to a desorption rate or uptake rate, but also to an equilibrium situation. As an example for soil contaminants, bioavailability has been used to indicate the fractions or amounts of contaminants that can desorb within a certain period of time [59,61,62], the amount of contaminants that can be taken up by organisms [56,63], but also the ratio between maximal desorption rate and maximal biodegradation rate [64]. Therefore, a clear definition of the term bioavailability should be given. In this thesis 'bioavailable' or 'bioavailability' refers to that fraction of the contaminants that can desorb or can be degraded within a specified period. In soil remediation practice this period will be determined by economic considerations.

Figure 1.2

Typical desorption curve of aged hydrophobic contaminants; in this example 10% of the contaminants exhibits slow desorption behaviour



PROCESSES DETERMINING RESIDUAL CONCENTRATIONS

More detailed studies of the desorption behaviour of hydrophobic compounds sorbed to soil showed that part of the contaminants desorb with a much lower rate than other part [49,65-67]. This results in a two-stage desorption behaviour: initially a stage during which desorption is relatively fast, followed by a stage with much lower desorption rates (see Figure 1.2).

Several mechanisms have been suggested to explain this two-stage desorption behaviour. Some studies showed that nano-sized pores in synthetic matrices led to very slow desorption of hydrophobic compounds [68,69]. In these studies it was hypothesised that desorption from nano-sized pores within the inorganic soil matrix accounted for the slow desorbing fraction of the contaminants. However, this contrasts the findings that low organic matter soils contain only small fractions of slowly desorbing contaminants [70].

Other studies kept the soil organic matter responsible for slow desorption. Several authors stated that soil organic matter consists of elastic and rigid regions, which were believed to resemble the rubbery and glassy state of synthetic polymers [66,71]. Diffusion and desorption of hydrophobic compounds takes place at a much lower rate from the rigid regions than from the elastic regions of the soil organic matter [66]. Further, it has been reported that these rigid regions exhibited non-linear sorption isotherms, which were explained by contaminant sorption in nano-pores in the rigid regions [72-74]. Desorption from these nano-pores is very slow. Some more recent studies doubted the existence of elastic and rigid regions in the soil organic matter [75]. An alternative

mechanism has been suggested: after sorption of contaminants in the soil organic matter, structural rearrangements of the soil organic matter lead to a physical incorporation of (part of) the contaminants within the soil organic matter structure [76,77]. Molecular modelling calculations showed that contaminant can indeed become entrapped within soil organic matter structures [78,79].
Besides natural organic matter, antropogenic high carbonaceous material, like soot and coal tar particles, can cause slow desorption of contaminants. This slow desorption is caused by very strong sorption and slow diffusion of the hydrophobic contaminants through these matrices [80-82].

OBJECTIVES AND OUTLINE OF THIS THESIS

Biological remediation of PAH contaminated soils has many advantages over physical and chemical remediation technologies. However, although most PAH compounds are readily degradable by micro-organisms, high residual concentrations in the soil often remain, because of very slow desorption of part of the contaminants. This hampers the implementation of bioremediation technologies for PAH polluted soils and sediments.
The general objective of this study is to investigate methods to improve bioremediation of PAH contaminated soils, this means to decrease residual concentrations after bioremediation. Because residual concentrations are determined by desorption kinetics of the contaminants from the soil, these methods are directed towards an increase of the desorption rate.
In Chapter 2 of this thesis, a literature survey is given on methods that might increase the desorption rate of hydrophobic compounds. Different methods are discussed and evaluated. The most promising methods are selected for further study in laboratory experiments.
The effects of two pre-treatment methods, short-term heating and soaking with an organic solvent, on the subsequent biodegradation process are studied in Chapter 3. Treatment temperatures and solvent concentrations are varied.
In Chapter 4, the optimal conditions for short-term heating as a pre-treatment step for bioremediation of contaminated soils are investigated. Desorption kinetics of several PAH compounds after pre-treatments at various temperatures, times and water contents are determined.
Biodegradation rates and desorption rates of PAH are compared for various contaminated soils in Chapter 5. Based on observations in this chapter, a model is

constructed that describes desorption and degradation of PAH and growth of PAH degrading biomass. This model and its validation with experimental results are described in Chapter 6.
The technical and economical possibilities for application of thermal pre-treatment in soil remediation practice are discussed in Chapter 7 of this thesis. Different techniques for thermal pre-treatment in several soil bioremediation processes are discussed and costs of the techniques are calculated. The costs of combined thermal pre-treatment and subsequent bioremediation are compared to costs of alternative strategies for remediation of soil.
In Chapter 8, this thesis is concluded with a discussion of the results presented in this thesis in relation to the application of a thermal pre-treatment in current soil remediation practice.

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2

Selection of Chemical and Physical methods to improve Bioremediation of HOC Contaminated Soils

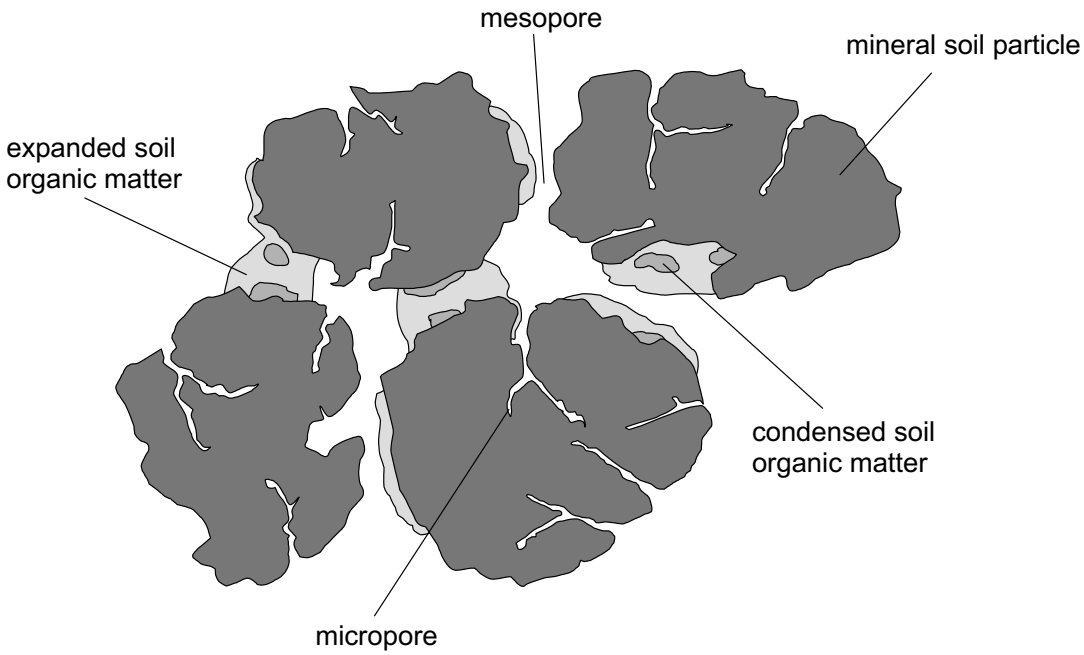
A literature survey is presented of methods that might increase the desorption rate of PAH and other hydrophobic compounds. Different methods were discussed and evaluated based on their proposed effects. It was found that almost all methods that tried to improve bioremediation by an increase of the pore diffusion rate led to higher initial remediation rates. However, these methods did not lead to lower residual concentrations. Only a redistribution of contaminants over the soil had proofed to decrease residual concentrations after biodegradation. Redistribution can be performed by soaking of the soil in an organic solvent. Also, an increase of the organic matter diffusivity showed good perspectives for decreasing residual concentrations, although literature data on this were scarce. A diffusivity increase can be performed by a short-term heating of the soil or by an extraction of multivalent cations from the soil. However the latter is a timely and expensive method. Soaking with an organic solvent and short-term heating were selected for further study.

INTRODUCTION

Biological remediation of soils and sediments that are contaminated with hydrophobic organic contaminants, like PAH, has not been successful, due to large fractions of the contaminants that are resistant for biodegradation. Generally, this resistance is not caused by poor biodegradability of the contaminants, as most contaminants are readily degraded when dissolved in water [1,2] or freshly spiked to soils or sediments [3-5]. Because most microorganisms can only degrade dissolved compounds [6-8], transport of the contaminants from the soil to the microorganisms determines the bioremediation results. To improve these bioremediation results, it is therefore necessary to find methods and techniques to increase the transport rate.

The transport rate of contaminants from the soil involves several processes. To discuss these processes and methods to increase the rate of these processes, a general representation of an aggregate in soils or sediments is shown in Figure 2.1.

Figure 2.1 Schematic representation of a soil aggregate



This figure shows a soil aggregate consisting of mineral particles (sand, silt, clay) and soil organic matter. Soil organic matter has been operationally classified in fulvic and humic acids and humin [9]. Hydrophobicity of the soil organic matter increases from fulvic acids towards humin.
The pores between the solid soil components (mineral and organic) are filled with water (slurry processes) or air and water (landfarming, in-situ processses). Pores can be classified in macropores (between soil aggregates, > 2 μm), mesopores (between mineral particles, 0.5-2 μm), and micropores (within clay domains and soil organic matter, < 0.5 μm). In slurry processes, no macropores can be defined, due to dispersion of the soil. Mesopores and micropores are generally to small for bacteria (size 1-2 μm). Consequently, contaminants have to desorb from soil aggregates to be biodegraded. The desorption rate can be determined by several processes.

Several studies have shown that the diffusion rate of contaminants through mesopores in soil can determine the desorption and bioremediation rate [10-12]. A decrease of the aggregate size will then lead to shorter diffusion lengths and, consequently, to faster remediation. Also an increase of the diffusion coefficient leads to improved remediation. Further, higher contaminant concentrations in the aqueous phase in the pores will lead to faster diffusion rates. The aqueous phase concentrations are determined by the sorption isotherms of the contaminants to the solid soil particles.
It has often been shown that contaminants mainly absorb in or adsorb to soil organic matter and not to mineral surfaces [13-15]. Remediation rates might then be increased by either a removal of (part of) the soil organic matter or a decrease of the sorption strength of the contaminants to the soil organic matter.
In contrast to pore diffusion, others have suggested that diffusion in soil organic matter itself limits removal of contaminants from soil [16,17]. In this case, a higher diffusivity of contaminants in the soil organic matter improves bioremediation.
Finally, it has been suggested that the high residual concentrations after bioremediation are caused by sorption of contaminants to places from which desorption is very slow. Both micropores within the inorganic soil matrix [6,18] and condensed or glassy regions in the soil organic matter [19,20] have been suggested to cause this slow desorption of contaminants. A redistribution of contaminants from places with slow desorption to places that exhibit faster desorption will then increase the bioremediation rate.

In this chapter, theoretical concepts and experimental results from literature are discussed for techniques and methods that might improve bioremediation by

- decrease of aggregate sizes
- increase of the diffusion coefficient for pore diffusion
- removal of soil organic matter
- decrease of the sorption strength to soil organic matter
- increase of organic matter diffusivity
- redistribution of contaminants to places that exhibit faster desorption

At the end, a selection is made from the discussed methods. The methods that show to be the most promising for reduction of residual concentrations of hydrophobic contaminants, especially PAH, will be further studied on their ability to improve bioremediation results.

PRE-TREATMENT METHODS

DECREASE OF SOIL AGGREGATE SIZE

When modelling soil processes, a soil aggregate can often be represented as a spherical particle with a homogenous pore distribution. The release of compounds from a spherical partical can be described with the radial diffusion equation (eq. 2.1). In case diffusion of contaminants through the soil mesopores determines the biodegradation rate and not release from soil organic matter or micropores, release of contaminants from a soil aggregate can then be described with this radial diffusion equation [10]:

$$\frac{\partial S}{\partial t} = D_{eff} \cdot \left(\frac{\partial^2 S}{\partial r^2} + \frac{2}{r} \frac{\partial S}{\partial r} \right) \qquad (2.1)$$

with the boundary conditions:

$$\left. \frac{\partial S}{\partial r} \right|_{r=0} = 0 \qquad (2.2)$$

$$S_{r=R} = 0 \qquad (2.3)$$

where, S is the concentration of the contaminant in a soil aggregate (mg/kg); D_{eff} is the effective diffusion coefficient of contaminant for diffusion through soil pores (m^2/s); t is the time (s); r is the distance from the centre of the soil aggregate (m); R is the radius of the soil aggregate (m).

With eq. 2.3 it is assumed that there will be no external mass transfer limitations. For the following initial condition, which means an initial homogeneous contaminant distribution over the soil aggregate:

$$S = S_0 \quad for: \quad 0 \le r < R \quad at: \quad t = 0 \tag{2.4}$$

eq. 2.1 can be integrated, which yields the following equation [21]:

$$\frac{S_t}{S_0} = \frac{6}{\pi^2} \sum_{n=1}^{\infty} \frac{1}{n^2} \cdot e^{-\left(\frac{n \cdot \pi}{R}\right)^2 D_{eff} \cdot t} \tag{2.5}$$

where, S_t is the average concentration in the soil aggregate at time t (mg/kg); S_0 is the initial concentration in a soil aggregate (mg/kg).
From eq. 2.5 it can be derived that either an increase of the effective diffusion coefficient or a decrease of the soil aggregate radius can increase the removal rate. A two-fold decrease of the radius leads to a four-fold increase of the removal rate.
A decrease of the soil aggregate size can be obtained by several methods. The most direct way to decrease the aggregate size is milling of the soil aggregates.
Steinberg et al. [18] reported an increase of desorption of the soil fumigant 1,2-dibromoethane after milling. However, this was attributed to a release of 1,2-dibromoethane which had been entrapped in soil micro-pores and not to a decrease of diffusion lengths. Release of this contaminant was found to be highly independent of the aggregate radius, determined for different size fractions of the unmilled soil. Ball and Roberts [22] studied the uptake of tetrachloroethylene and tetrachlorobenzene, and they found that milling increased the uptake rate of these compounds. However, this increase was much smaller than could be expected from the radial diffusion model (eqs. 2.1-2.5). Both results suggest that the appropriate diffusion length is not the soil aggregate radius, but that diffusion takes place at a much smaller scale, e.g. organic matter domains within a soil aggregate. No effect of milling was found by Carroll et al. [16] who studied PCB desorption from sediment. This sediment contained a much higher organic matter content than the soils in the two previous studies, which further suggests that not diffusion through soil meso-pores is limiting removal but release from soil organic matter.
Another method to decrease the soil particle size is the use of ultrasonic waves. Ultrasound is widely used in industry for the dispersion of solids and liquids [23]. It can also be used for the disruption of soil aggregates and the dispersion of

these aggregates. Hatzinger and Alexander [5] found that ultrasonic disruption of soil aggregates increased the phenanthrene biodegradation rate, for both a fresh and an aged contamination. The extents of degradation for disrupted and intact soils were similar, however. Also Gauger [24] found no effect of ultrasonic treatment on the extent of bioremediation of PAH contaminants in a gas plant soil.

INCREASE OF THE DIFFUSION COEFFICIENT

From the equations of the radial diffusion model (eqs. 2.1-2.5) can be seen that the rate at which contaminants are released from a soil aggregate is determined by both the aggregate size and the effective diffusion coefficient. The effect of the aggregate size is discussed in the previous paragraph. The consequences for the contaminant release rate of changing the effective diffusion coefficient will be discussed in the next praragraphs. The effective diffusion coefficient can be derived from the diffusion coefficient of the contaminant in water and the sorption isotherm of the contaminant [11]. In case of a linear sorption isotherm, the following equation for D_{eff} can be obtained:

$$D_{eff} = \frac{D_b \cdot \mathrm{f}(\tau, c) \cdot \varepsilon}{\varepsilon + (1-\varepsilon) \cdot \rho \cdot K_p} \qquad (2.6)$$

where, D_b is the diffusion coefficient of the contaminant in water (m^2/s); ε is the porosity of the soil particle (-); ρ is the density of the soil matrix (kg/m^3); K_p is soil-water partition coefficient (m^3/kg). The effective diffusion coefficient is corrected for tortuosity (τ) and constrictivity (c) effects by f(τ,c).
From eq. 2.6 it can be derived that both an increase of the diffusion coefficient in water and a decrease of the soil-water partition coefficient can increase the effective diffusion coefficient. The porosity, density, tortuosity and constrictivity of the soil matrix are very hard to change.
The diffusion coefficient of compounds in water depends on the temperature [25]. Table 2.1 shows the diffusion coefficient of phenanthrene (a 3-ring PAH compound) for various temperatures. Table 2.1 shows that the increase of D_b due to a temperature rise is relatively small, only 20 % for every 10-degree increase. Because degradation of most contaminants can only take place in the mesophilic range (20-40 °C) [26], a temperature rise can only be small and, consequently, the increase of the diffusion coefficient will be small too.

Table 2.1

Diffusion coefficients and partition coefficients of phenanthrene in water for various temperatures

T (°C)	D_b (m^2/s)[a]	K_{om} (m^3/kg)[b]
20	$0.62 \cdot 10^{-9}$	6.03
30	$0.80 \cdot 10^{-9}$	4.72
40	$1.01 \cdot 10^{-9}$	3.71
50	$1.25 \cdot 10^{-9}$	3.02
60	$1.51 \cdot 10^{-9}$	2.46

[a]calculated from [25]; [b]calculated from [14,27]

DECREASE OF THE SOIL ORGANIC MATTER CONTENT

Equation 2.6 showed that the effective diffusion coefficient can be increased by an increase of the diffusion coefficient of the contaminant in water and a decrease the soil-water partition coefficient. Methods to decrease the soil-water partition coefficient will be discussed in the following two paragraphs. The soil-water partition coefficient is mostly dependent on the organic matter content and the partition coefficient between the soil organic matter and water [27,28]:

$$K_p = f_{om} \cdot K_{om} \tag{2.7}$$

where, f_{om} is the organic matter content of the soil (-); K_{om} is the organic matter-water partition coefficient (m^3/kg).
It is clear from eq. 2.7 that removal of (part of) the soil organic matter leads to a decrease of the partition coefficient. The humic and fulvic acids, that are part of the soil organic matter, can be removed by a treatment of the soil with caustic solutions [9]. Carroll at al. [16] found an increase of the initial desorption rate of PCBs from sediment after treatment with a sodium hydroxide solution. The residual fractions after desorption were similar for treated and non-treated soil, however. The humin fraction of the soil organic matter can not be removed by a caustic treatment. This fraction is the most hydrophobic part of the soil organic matter and, therefore, sorption to this fraction is the strongest. This suggests that release of the contaminants from this humin fraction determines the extent of bioremediation.

DECREASE OF THE ORGANIC MATTER-WATER PARTITION COEFFICIENT

The soil-water partition coefficient is also dependent on the organic matter-water partition coefficient, which can be seen from eq. 2.7. In this paragraph, four

different methods will be discussed to change the organic matter-water partition coefficient.
First, the organic matter-water partition coefficient decreases when the temperature increases, as sorption of hydrophobic compounds to soil organic matter is an exothermic process [29,30]. The following equation can be derived for the partition coefficient as a function of temperature [27]:

$$\ln\left(\frac{K_{om,T_1}}{K_{om,T_2}}\right) = \frac{\Delta H_s}{R}\left(\frac{1}{T_1} - \frac{1}{T_2}\right) \qquad (2.8)$$

where, ΔH_s is the enthalpy for solution of the contaminant in water (J/mole); R is the gas constant ($J \cdot mole^{-1} \cdot K^{-1}$).
In Table 2.1 the partition coefficients of phenanthrene in a soil with 1 % organic matter for several temperatures are shown. This table shows that every 10-degree temperature increase leads to a 20-30 % decrease of K_{om}, which is relatively small. However, because D_b increases with increasing temperature (see Table 2.1), the combined effect of D_b increase and K_{om} decrease might lead to faster bioremediation. Maliszewska-Kordybach [31] found that a temperature increase from 10 to 25 °C indeed led to an increase of the degradation rate for spiked PAH contaminants. Cornelissen et al. [32] studied the effect of temperature on desorption of aged contaminants. They found that higher temperatures (60 °C compared to 25 °C) led to faster desorption rates of PCB, but that a fraction of the contaminants was resistant for desorption. This resistant fraction was not changed by the temperature increase.
Another way to decrease the organic matter-water partition coefficient is the use of surfactants. Surfactants increase the apparent water solubility of hydrophobic compounds by the formation of micelles in which these hydrophobic compounds can dissolve [33,34]. The following equation clearly shows that surfactants decrease the apparent value of K_{om} for surfactant concentrations above the critical micelle concentration (CMC):

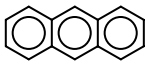
$$K_{om,app} = \frac{K_{om}}{1 + K_{mic} \cdot (C_{surf} - CMC)} \qquad (2.9)$$

where, $K_{om,app}$ is apparent organic matter-water partition coefficient in the presence of surfactants (m^3/kg); K_{mic} is the partition constant between micelles and the water phase (m^3/kg); C_{surf} is the surfactant concentration in the water

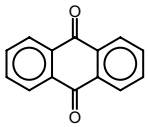
phase (kg/m^3); CMC is the critical micelle concentration, above this concentration surfactants will form micelles (kg/m^3).
On the other hand, the effect of the decreased partition coefficient is partly undone by lower diffusion coefficients of the micelles compared to the diffusion coefficients of the individual contaminants molecules in the aqueous phase [35]. The overall effect, however, shows an increase of the effective diffusion coefficient when surfactants are applied. The effect of surfactants on bioremediation has been extensively studied [35-38]. It was found that surfactants are able to improve desorption and biodegradation rates. No information could be found of the effects of surfactants on the extent of bioremediation. On the other hand, the use of surfactant could also lead to a retarded bioremediation due to toxic effects of the surfactants for microorganisms [36], or oxygen and nutrient depletion caused by biodegradation of the surfactants [39].
As an alternative for surfactants, natural dissolved organic matter (DOM) can be used to decrease the organic matter-water partition coefficient. This DOM consists of dissolved humic and fulvic acids. It is very resistant to biodegradation and generally not toxic for microorganisms. It has been found that humic and fulvic acids can also form micelles, similar to surfactants [40,41]. Several studies have shown that DOM increases the apparent solubility of hydrophobic compounds and facilitates transport through soil [42-44].
Finally, the organic-water partition coefficient can be decreased by a partial oxidation of the contaminants. The addition of hydrophilic groups (e.g. oxide and hydroxide groups) to hydrophobic compounds increases the solubility of these compounds; as an example Figure 2.2 shows some reaction products of the oxidation of anthracene and their solubilities in water [45]. Several authors have found that spiked contaminants are readily oxidised with radical oxidants [46-48]; oxidation products could be readily biodegraded.
Cuypers et al. [49] compared results from biodegradation experiments with oxidation experiments for several soils and sediments with aged PAH contaminations. In the oxidation experiments, persulfate was used as oxidising agent. They found that biodegradation and oxidation removed the same portion of the PAH from the soil. These results suggest that oxidation can not cope with the recalcitrant fraction of the contaminants and can not be used to reduce residual concentrations.
Another disadvantage of partial oxidation can be the formation of toxic oxidation products that inhibit biodegradation [50,51].

Figure 2.2

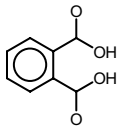
Oxidation products of anthracene and their solubilities after oxidation with hydroxide radicals [45]



anthracene
solubility = 70 μg/l



anthraquinone
solubility = 600 μg/l



phtalic acid
solubility = 7 g/l

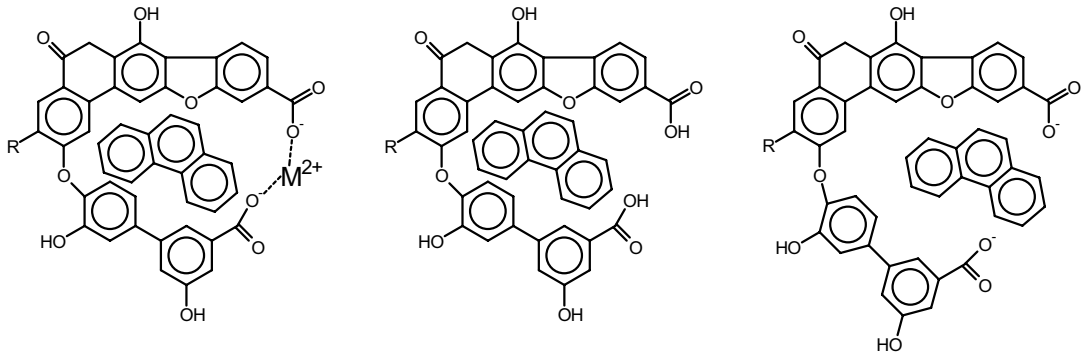
INCREASE OF SOIL ORGANIC MATTER DIFFUSIVITY

As mentioned in the introduction, bioremediation of contaminated soils and sediments might not only be limited by diffusion through the soil pores, but also by diffusion through the soil organic matter [16,17]. The removal rate of the contaminants can then be increased by an increase of the diffusivity of the soil organic matter.

Several authors have shown that the ionic strength and pH of the water phase affect the sorption of hydrophobic compounds to dissolved organic matter [52-54]. Metal ions (especially multivalent ions) and protons can adsorb to reactive groups of the organic matter (e.g. COO^-), thereby neutralising the mutual repulsion of these groups. As a result, the organic matter will have a more condensed structure, which obstructs desorption of sorbed organic contaminants. Figure 2.3 illustrates how metal ions and protons can affect desorption of phenanthrene, a 3 ring PAH compound.

Extraction of multivalent cations from soil or change of ionic strength of the waterphase might then increase removal rate of organic contaminants. Hall et al. [55] studied the effect of sea salt concentrations on desorption and transport of spiked contaminants from soil. They found a very small increase of atrazine removal at high salinity (5 ppt compared to 25 ppt). Aged contaminants were studied by Hegemann et al. [56], who found a very small effect of salinity on phenanthrene and benzo[a]pyrene sorption. These results contrast with the results found for sorption of hydrophobic compounds to dissolved organic matter mentioned earlier. This might be explained by the fact that soil organic matter contains less reactive groups than dissolved organic matter [9], and consequently ion concentrations will have a smaller effect on soil organic matter structure.

Figure 2.3 Effects of (multivalent) cations and protons on organic matter structure and on binding of hydrophobic compounds by soil organic matter (modified from [52] and [54])



No literature was found on the effect of metal ion extraction on bioremediation. However, it has been shown that removal of metals from soil is a prolonged and expensive process, using large amounts of acid or extracting agents [57].
A brief change of the soil temperature can also alter the structure of soil organic matter. Several authors have proposed that soil organic matter consist of glassy and rubbery regions [58,59], analogous to glassy and rubbery phases of synthetic polymers [60]. Sorption of contaminants to the glassy regions accounts for the non-available portion of hydrophobic organic contaminants, as diffusion of organic compounds through glassy polymers is much slower than through rubbery polymers [16]. Xing and Pignatello [61] showed that sorption isotherms became increasingly linear with increasing temperature and that sorption of the contaminants became weaker. This was attributed to relaxation of the glassy regions and a conversion of the glassy structure to a rubbery one. LeBoeuf and Weber [19] also detected a glassy-rubbery transition after a temperature increase in commercial humic acids. In case these conversed glassy regions remain in a rubbery state after returning to normal temperatures, availability of the sorbed contaminants has increased.

As mentioned in the introduction, it has often been shown that freshly spiked contaminants are readily biodegraded whereas aged contaminants exhibit high residual concentrations with low desorption and biodegradation rates [3-5]. This is generally attributed to slow diffusion of contaminants into regions that exhibit strong sorption and slow desorption, like glassy regions of soil organic matter [19,20] or micro-pores [6,18]. A redistribution of these contaminants to regions from where fast desorption is possible will lead better bioremediation results. Addition of a small amount of organic solvent to soil increases the solubility of the contaminants [62,63]. As a result, more contaminants tend to dissolve in the liquid phase. After evaporation of the solvent, the contaminants will sorb in a region that exhibits high desorption rates, similar to a freshly spiked contamination. Field et al. [64] showed that, after soaking of a PAH contaminated soil for 2 days with acetone, both biodegradation rates increased and residual concentrations decreased. This was found for biodegradation by both white rot fungi and endogenous bacteria.

DISCUSSION

The results of the previous discussion of methods to improve biodegradation of hydrophobic organic compounds has been summarised in Table 2.2. This table shows the effect on the initial degradation rates and the residual concentrations after bioremediation.

Table 2.2 clearly shows that almost all methods which tried to improve bioremediation by an increase of the pore diffusion rate (milling - partial oxidation) led to higher initial remediation rates. This suggests that for the first stage of a bioremediation process, the removal rate of the contaminants is determined by diffusion through soil pores. Also others have stated that diffusion limits initial bioremediation.

On the other hand, these methods did not lead to lower residual concentrations. The slow removal of the residual fractions should then be caused by other mechanisms than mesopore diffusion. Diffusion through soil organic matter or desorption of contaminants from glassy organic matter or from micropores are the most obvious mechanisms. Diffusion through soil organic matter might be improved by extraction of metals ions, a change of ionic strength and pH, and a thermal pre-treatment. Unfortunately, literature data for these methods are scarce. A change of ionic strength or pH will probably be the least successful, because

Table 2.2 Summary of effects of methods to improve bioremediation of hydrophobic contaminants

principle	*method*	*degradation rate*[a]	*residual concentration*[a]
decrease of aggregate size	milling	+	-
	ultrasonic waves	+	-
increase of D_b	temperature increase	+	-
removal of soil organic matter	caustic extraction	+	-
decrease of K_{om}	temperature increase	+	-
	surfactants	+/-	n.d.
	dissolved organic matter	n.d.	n.d.
	partial oxidation	+	-
increase of soil organic matter diffusivity	cation extraction	+/-	n.d.
	thermal pretreatment	n.d.	n.d.
redistribution of contaminants	soaking with solvent	+	+

[a] '+' means increase of initial degradation rate or decrease of residual concentrations; '-' means no effect or a negative effect on degradation rate or residual concentrations; 'n.d'. means no data found in literature

others already found very little effect on the desorption rates of aged contaminations. The effects of an extraction of metals on biodegradation are unknown, but the extraction process is very timely and expensive and therefore not suitable as a pre-treatment method [57]. The effects of a brief thermal pretreatment on bioremediation are unknown, but it is speculated that a thermal pretreatment leads to an improved biodegradation process.

The incorporation of contaminants in glassy organic matter or micropores can be undone by a redistribution of the contaminants among the soil particles. This redistribution can be brought about by soaking of the contaminated soil with an organic solvent. Field et al. [64] found for one soil that soaking with acetone decreased residual concentrations.

The most promising methods to improve bioremediation of soils contaminated with hydrophobic organic compounds are a thermal pretreatment and soaking with an organic solvent. In Chapter 3, experimental results of the effects of these two methods on biodegradation are shown.

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3

Enhancement of PAH Biodegradation in Soil by Physical/Chemical Pretreatments

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The effects were studied of short-term heating of contaminated soil and soaking in an organic solvent on the subsequent biodegradation of PAH. In a clayey soil washing sludge with a high organic-matter content (12 %), heating at 120 °C for one hour increased the degree of degradation after 21 days of an aged PAH contamination from 9.5 ± 0.7 % to 27 ± 5 %. Lower temperatures resulted in smaller increases. The observed increase in biodegradation is caused by either transfer of PAH from sorption sites with low desorption rates to those with high ones or transformation of slow-sorption sites into fast-sorption sites. Soaking of the above sludge in a 4:1 ($^{v}/_{v}$) acetone-water mixture increased the degree of degradation from 9.5 ± 0.7 % to 20.4 ± 1.4 %, probably as a result of dissolution of the PAH in the pore liquid during soaking. Thermal pretreatment of a contaminated sandy soil with a low organic-matter content showed no significant effect on the degradation of aged PAH. Soaking of the sandy soil increased the degradation of only PAH of high molecular weight, namely from 24 ± 5 % to 48 ± 7 %.

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INTRODUCTION

Biological decontamination of soils polluted with hydrophobic organic compounds (PAH, HCH, oil, etc.) has not always been successful due to low degradation rates and high residual concentrations that do not meet the legal clean-up guidelines [1,2]. This problem, also called "limited bioavailability", is caused mainly by a low rate of desorption of contaminants from soil particles and is, in general, not due to slow degradation by micro-organisms. Two main reasons for this slow desorption are given in literature: first, slow diffusion of contaminants through the pore liquid due to sorption to soil organic matter [3-5], and second slow diffusion of contaminants through soil organic matter [6,7]. According to several authors, desorption occurs in two phases: a phase of fast desorption, followed by one of slow desorption [6-9]. Regarding retarded pore diffusion, the two desorption phases may be the result of desorption from pores of different size: macro-pores and micro-pores [4]. Regarding diffusion through soil organic matter, the two desorption phases have been attributed to diffusion through expanded and condensed regions in the soil organic matter [6,10,11]. Furthermore, recent literature shows that physical incorporation into micro-voids in condensed soil organic matter may also cause slow desorption from soil organic matter [9].

The objective of the present study was to investigate the possibilities for enhancing the bioavailability of hydrophobic organic contaminants in soil by subjecting the soil to two different short-term pretreatments: raising the soil temperature, and soaking the polluted soil in a water-miscible organic solvent.
It was demonstrated that both an increase in temperature [12,13] and the addition of an organic solvent [14] increase the rate of mass transfer of hydrophobic compounds within soil particles. Such an increase in the rate of mass transfer may lead to redistribution of contaminants from sites exhibiting a slow desorption rate to those exhibiting a fast one.
It was also demonstrated that both higher temperatures and organic solvents affect the structure of soil organic matter in such a way that desorption is facilitated [9,10]. If such a change in structure is irreversible or slowly reversible, the bioavailability of the hydrophobic compounds will increase.

MATERIAL AND METHODS

SOILS

Two different types of soil were used. The first was a sludge residue obtained from a soil washing plant. The original soil had been obtained from a former gas plant site in Kralingen, the Netherlands. The soil had been separated at a cut-off diameter of 63 μm using hydrocyclones. The sludge residue - i.e. the product containing the smallest particles - was air-dried and sieved at 2 mm to remove coarse, light material that could not be separated by hydrocyclonage. The total PAH concentration (16 EPA) was 115 mg/kg. The soil also contained 300 mg/kg mineral oil and 9 mg/kg cyanide.
The other type of soil used was a sandy soil from a wood preservation plant in Schijndel, the Netherlands. This soil was air-dried and sieved at 2 mm to remove small stones, grass leaves, etc. The total PAH concentration (16 EPA) was 101 mg/kg.
The organic matter contents and mineral compositions of the soils are given in Table 3.1.

INOCULUM

To promote biodegradation after pretreatment, an enrichment culture was prepared from a slurry consisting of 100 ml of mineral medium, 0.2 g of yeast extract (Oxoid) and 10 g of harbour sludge from the 1st Petroleum Harbour, Amsterdam. The dry matter content of the harbour sludge was 45%. The dry matter of the sludge had an organic matter content of 9.9%. The total PAH concentration (16 EPA) was 5,400 mg/kg. The sludge also contained 20,000 mg/kg mineral oil.
The mineral medium contained 0.2 g of NH_4NO_3, 0.1 g of $MgSO_4 \cdot 7H_2O$, 0.15 g of $CaCl_2 \cdot 6H_2O$, 20 mg of KH_2PO_4, 80 mg of K_2HPO_4 and 5 mg of $FeCl_3$ per litre of medium. All chemicals were obtained from Merck. The slurry was incubated in a 300 ml Erlenmeyer flask at 30 °C for two weeks on a shaker operating at 130 rpm. Before using the resultant enrichment culture as an inoculum, its PAH concentrations were determined. Samples (20 ml) were taken in triplicate and centrifuged at 10,000 rpm for 10 minutes. The residue was extracted with 15 ml of 1-methyl-2-pyrrolidinone (99 %, Acros) in a microwave oven (MDS 2100, CEM corporation) at 130 °C for one hour, as described previously [15]. After extraction, the 1-methyl-2-pyrrolidinone was centrifuged at 10,000 rpm for 10 minutes and the supernatant was analysed by HPLC.

Table 3.1

Composition of the soils used in experiments

soil	*org. matter*[a]	*clay*[b]	*silt*[b]	*sand*[b]
Kralingen	12.3	27	46	27
Schijndel	1.0	3	20	77

[a] in percentage of dry weight determined after heating at 500 °C, [b] in percentage of the mineral part

THERMAL PRETREATMENT

2.5 ml of the above-mentioned mineral medium was added to 60-ml serum flasks containing 5 g of soil. Next, thermal pretreatment was carried out as follows. Some closed flasks were placed in a water bath at 70 °C and others at 95 °C. After 1 hour, the flasks were removed and cooled under running water. Yet other closed flasks were placed in an autoclave and allowed to stand for 1 hour at 120 °C. After cooling to 100 °C in the autoclave, the flasks were cooled further under running water.

SOAKING

5 g of soil was added to 60-ml serum bottles together with 1.25 ml or 2.5 ml of an acetone/water mixture (4:1 $^{v}/_{v}$). After 2.5 ml of the acetone/water mixture was added, the soil was just below the solvent level. After 24 hours, the acetone and water were evaporated at 40 °C in 24 hours.

MINERALISATION

To determine the mineralisation rate of the PAH compounds, mineral medium was added to the serum flasks to a total liquid volume of 7.5 ml. Next, the enrichment culture (2.5 ml) was added to each flask. The flasks were mixed at 30 °C on an end-over-end shaker at 22 rpm. On days 0, 3, 7, 11, 16 and 21, flasks were removed from the shaker and sacrificed for analysis. To verify that the oxygen content was not limiting biodegradation, the oxygen and carbon-dioxide concentrations in the flasks were determined by GC. To extract the PAH from the soil, 40 ml of acetone was added to each flask and the flasks were treated ultrasonically in a bath (Retsch UR2) for 15 minutes. Then, the flasks were

shaken end-over-end for 1 hour. After 1 hour, the flasks were removed from the shaker and 1.5 ml of the acetone was removed by centrifuging at 10,000 rpm for 3 minutes and analysed by HPLC. All experiments were carried out in triplicate. The extraction procedure with acetone as described above had earlier proved to be very efficient and effective [15].

ANALYTICAL EQUIPMENT

Oxygen and carbon dioxide were analysed using an Interscience 8340 GC with a Teflon column (1.5 m × 2 mm L × I.D.) packed with Chromsorb 108 (60-80 mesh) parallel with a steel column (1.2 m × 2 mm L × I.D.) packed with a 5 Å mole sieve (60-80 mesh), with a 1:1 split and a thermal conductivity detector. Helium was used as a carrier gas at a flow rate of 45 ml per minute. The temperatures were 110°C for the injector, 40°C for the oven, and 99°C for the detector.
The PAH extracts were analysed using the following HPLC system [15]: a GasTorr GT-103 degassing device, a Gynkotech 480 HPLC pump, a Spark Holland Basic-Marathon Autosampler, and a Waters 991 photodiode array detector. A Vydac 5 C18 reverse phase column (250 mm × 4.6 mm L × I.D.) with an external guard column and a solvent gradient program with acetonitrile (LabScan, HPLC-grade) and distilled water were used to separate the PAH. Concentrations were determined by UV absorbance at 254, 264, 287 and 335 nm.

RESULTS

Analysis of the PAH concentrations in the inoculum revealed that the inoculum was capable of degrading all (16-EPA) PAH. However, the degradation efficiencies decreased with increasing molecular weight and increasing hydrophobicity of the PAH. Earlier experiments showed that with the experimental set-up used spiked PAH could be completely degraded, which means that microbial constraints did not play any role.

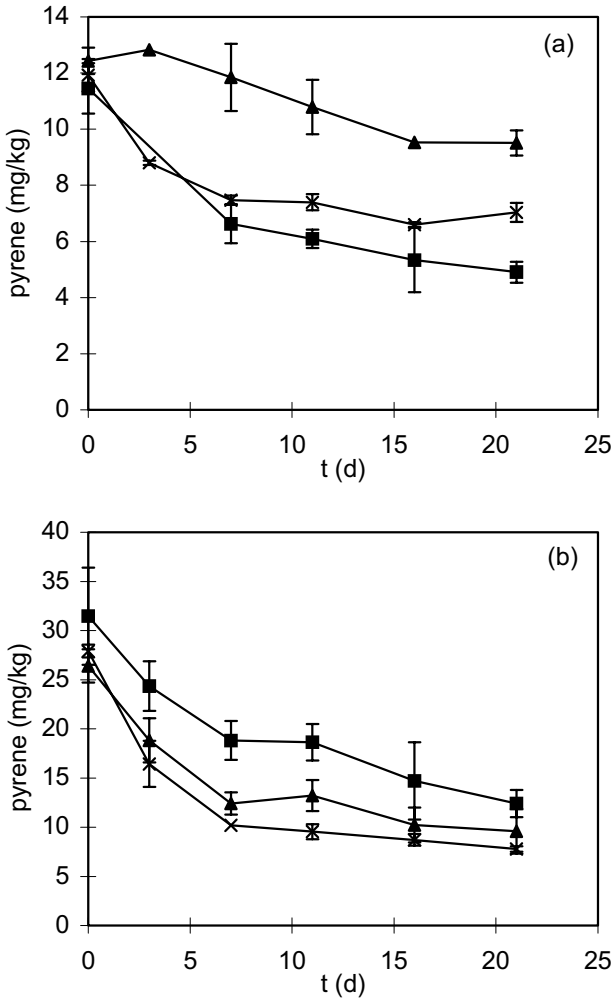
THERMAL PRETREATMENT

Figures 3.1a and 3.1b show degradation curves for pyrene in soils from Kralingen and Schijndel without pretreatment and after thermal treatment at 120°C. These curves are typical of 2-3 ring and 4-ring PAH. From Figure 3.1a, it can be derived that thermal pretreatment of soil from Kralingen at 120 °C for 1 hour increased the degradation rate and decreased the residual concentrations,

whereas thermal treatment of soil from Schijndel (Figure 3.1b) did not enhance the biodegradation.
Figures 3.2a and 3.2b show the percentages of 2-3 ring, 4-ring and 5-6 ring PAH degraded in soils from Kralingen and Schijndel after thermal pretreatment and 21 days of biological treatment. Asterisks indicate significant increases in degradation efficiency (Student t-test, 2-sided, 95 %) compared to the experiment during which no pretreatment was applied. 5-6 ring PAH were not degraded in soil from Kralingen. Thermal pretreatment at 95 °C and 120 °C increased the degradation efficiencies for 2-3 ring PAH from 17 % to 32 % and 54 %, respectively. At these temperatures, the degradation efficiencies for 4-ring PAH increased from 13 % to 17 % and 29 %, respectively. The overall PAH degradation efficiency increased from 10 % to 16 % and 27 % at 95 °C and 120 °C, respectively. A higher temperature led to significantly sharper decreases in residual concentrations.

Figure 3.1

Degradation of pyrene without pretreatment (▲), after thermal treatment at 120 °C for 1 hour (■) and after soaking with an acetone-water mixture (4:1 $^{v}/_{v}$) for 24 hours (×) in
(a) Kralingen soil
(b) Schijndel soil.
Error bars indicate one standard deviation.



Pretreatment at 70 °C showed no effect on residual PAH concentrations. Thermal pretreatment of soil from Schijndel had no significant effect on the PAH concentrations remaining after degradation (Student t-test, 2-sided, 95 %).

SOAKING

Figures 3.1a and 3.1b show degradation curves for pyrene in soils from Kralingen and Schijndel for a situation in which no pretreatment was applied and for one in which the soils were soaked with an acetone-water mixture (4:1 $^{v}/_{v}$) for 24 hours at a solvent concentration of 0.5 ml/g of dry soil. Figure 3.1a shows that in soil from Kralingen soaking increased the degradation rate and decreased the residual concentration for pyrene. However, the residual concentration was higher compared to that found after biodegradation and thermal pretreatment at 120 °C.

Figure 3.2

Percentages of PAH removal due to biological treatment during 21 days, after thermal pretreatment for 1 hour or after soaking with an acetone/water-mixture (4:1 $^{v}/_{v}$) for 24 hours in
(a) Kralingen soil
(b) Schijndel soil.
Error bars indicate one standard deviation. Asterisks indicate significantly different degradation percentages compared to non-pretreated samples (Student-t test, 2-sided, 95 %).

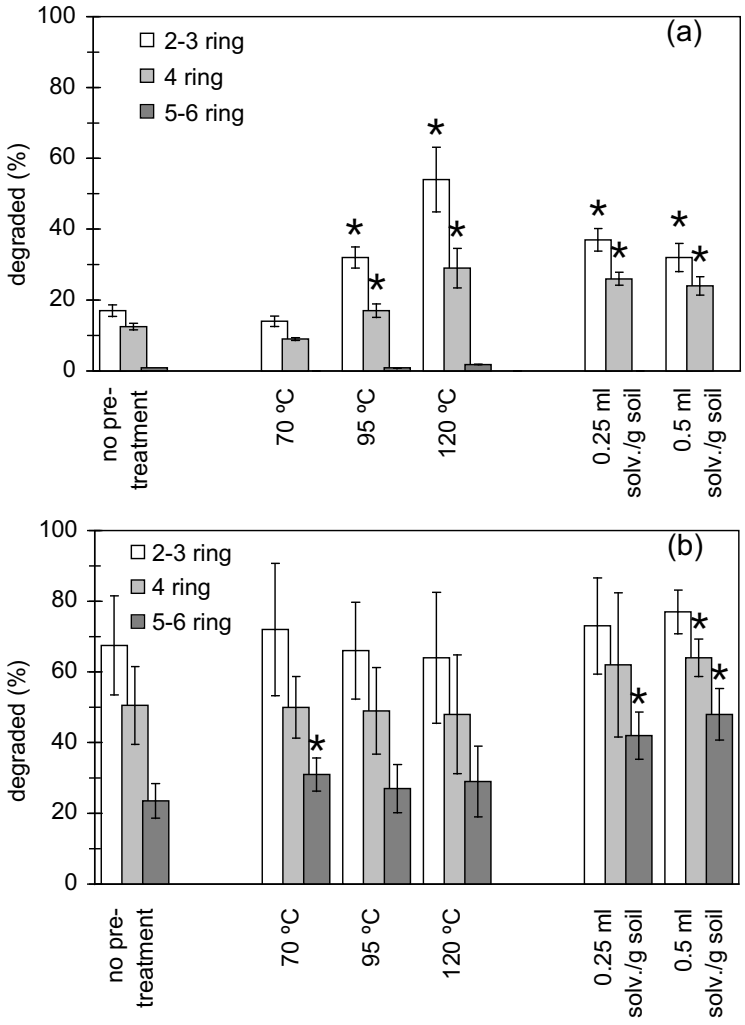


Figure 3.1b shows that soaking hardly increased the degradation rate for pyrene in soil from Schijndel.
Figures 3.2a and 3.2b show the percentages of 2-3 ring, 4-ring and 5-6 ring PAH compounds degraded after 21 days of biodegradation in soils from Kralingen and Schijndel after soaking. For soil from Kralingen, soaking with 0.25 ml and 0.5 ml of solvent/g of dry soil increased the degradation of 2-3 ring PAH from 17 % to 37 % and 32 %, respectively. The degradation efficiencies of 4-ring PAH increased from 13 % to 26 % and 24 %, respectively. The overall PAH degradation efficiency increased from 10 % to 20 % and 17 % for 0.25 ml and 0.5 ml of solvent/g of dry soil, respectively. The differences in degradation efficiency between the two solvent concentrations were not significant (Student t-test, 2-sided, 95 %).
Although soaking also decreased the residual concentrations in soil from Schijndel, these decreases were significant only for the degradation of 4-ring PAH for 0.5 ml of solvent per gram of soil and for the degradation of 5-6 ring PAH for both solvent-soil ratios (Student t-test, 2-sided, 95 %).

DISCUSSION

SOIL FROM KRALINGEN VERSUS THAT FROM SCHIJNDEL.

The results show that thermal pretreatment and soaking with an organic solvent increased the rate of biodegradation in soil from Kralingen but not in soil from Schijndel. This difference in pretreatment efficiency between the two soils can be explained in several ways. First, the two soils differ in structure and composition. The soil from Kralingen has high clay and organic-matter content values and therefore has a high internal porosity and a high sorption capacity inside the soil pores. The soil from Schijndel, on the other hand, is sandy, has a low organic-matter content and contains coal-tar particles. In this soil, sorption probably takes place into such particles. Thermal treatment may have a stronger effect on PAH contaminants sorbed onto or into soil organic matter than on those sorbed into tar particles. Hypothetical mechanisms accounting for the effect of pretreatment on contaminants sorbed onto soil organic matter are discussed below. Furthermore, soil from Schijndel shows a higher degradation efficiency for situations in which no pretreatment is applied, and it shows higher standard deviations for PAH concentrations (10-12 %) compared with soil from Kralingen (standard deviations: 5-6 %). Because of the high degradation efficiencies and standard deviations shown by soil from Schijndel, a possible small effect of pretreatment

may go unnoticed (especially for the degradation of 2-3 and 4-ring PAH after soaking).

THERMAL TREATMENT

There are a few mechanisms that may account for the increase in bioavailability resulting from thermal pretreatment.

First, an increase in temperature causes a decrease in soil-water partition coefficient, which expresses the distribution of contaminants between the soil phase and the surrounding water phase. Consequently, higher temperatures result in the dissolution of more contaminants. If most of the contaminants dissolve during thermal treatment, after having cooled down a situation may arise comparable to a spiked contamination. Spiked contaminants are almost completely biodegradable [16]. The partition coefficient of PAH decreases with 20-30 % for every 10-degree increase in temperature between 5 °C and 45 °C [17-19]. No data were found for higher temperatures. Assuming that the data found can be extrapolated, the partition coefficient will be at most 30 times lower at 120 °C compared to the situation existing at 20 °C. This is still far too low for desorption of most of the sorbed PAH. It can therefore be concluded that a decrease in partition coefficient during thermal treatment hardly effects the bioavailability of PAH directly.

Furthermore, the mass transfer within a soil particle increases with increasing temperature. This means that a raise in temperature can cause redistribution of contaminants between sorption sites differing in desorption rates. If a net transfer of PAH occurs from sites with a low desorption rate to those with a high rate, the bioavailability of these PAH will increase. Concerning retarded pore diffusion as a rate-limiting mechanism, the mass transfer depends on the effective diffusion coefficient, which is proportional to the diffusion coefficient in water and inversely proportional to the partition coefficient [3-5]. The diffusion coefficient in water increases 4 to 5 times with an increase in temperature from 20 °C to 120 °C [20]. This means that the effective diffusion coefficient increases at most 150 times with an increase in temperature from 20 °C to 120 °C.

Concerning intra-organic matter diffusion as a rate-limiting mechanism, the diffusion coefficient of polymeric materials can give an indication of the diffusion rate occurring in soil organic matter. For polymer diffusion, activation energies ranging from 35 kJ/mole to more than 100 kJ/mole were reported. However, most of the activation energies reported are within the range of 40-70 kJ/mole [12,21], which means that the diffusion coefficient increases 65 to 1,500 times with an increase in temperature from 20 °C to 120 °C. Several authors determined activation energies for the desorption of hydrophobic compounds

from soils and sediments. Energies ranging from 46 kJ/mole to 66 kJ/mole [13] were found, which means that the desorption rates increased between 120 and 1,000 times with an increase in temperature from 20 °C to 120 °C.
All these figures regarding the effect of higher temperatures on effective diffusion coefficients, polymer diffusion coefficients and desorption rates show that mass transfer strongly increases with an increase in temperature and that redistribution can occur between different sorption sites kinetically.
Thermodynamically, redistribution during heating will occur only if there is a positive enthalpy for the transfer of contaminants from slow-sorption sites to high-sorption ones. Cornelissen et al. [13] reported enthalpies ranging from 10 kJ/mole to 15 kJ/mole for PAH compounds in sediments. This means that, as a result of an increase in temperature from 20 °C to 120 °C, the fraction of contaminants sorbed at fast sites increased 2 to 3 times, which is comparable with the experimental results.
From another point of view, this enthalpy for contaminant transfer may be also that for the transformation of slow-sorption sites into fast-sorption sites, which may also explain the increase in bioavailability. Xing and Pignatello [9] and LeBoeuf and Weber [10] found that an increase in temperature caused sorption isotherms to become more linear, which was attributed to the elimination of micro-voids as a result of conversion of condensed soil organic matter into soil organic matter with a swollen structure. They stated that physical incorporation of hydrophobic compounds into such voids in condensed soil organic matter limited biodegradation. If such elimination of micro-voids is irreversible or slowly reversible, the bioavailability of the hydrophobic compounds will increase after thermal treatment.

SOAKING WITH AN ORGANIC SOLVENT

The most prominent effect of soaking with an organic solvent is a change in the partition coefficient. The soil-solvent partition coefficient decreases exponentially with an increase in acetone concentration [22,23]. In a 4:1 acetone-water mixture, the partition coefficient decreases with a factor ranging from 10^4 to 10^6. This means that almost all PAH dissolve in the liquid phase during soaking. Using several soils and sediments, Noordkamp et al. [15] demonstrated that a 4:1 acetone-water mixture can desorb more than 95 % of all the PAH present within one hour. Resorption of PAH after evaporation of the acetone may lead to a situation comparable to a spiked contamination, which is almost completely biodegradable. Surprisingly, the results of our experiments show that soaking resulted in only a minor increase in bioavailability.

Furthermore, Xing and Pignatello [9] demonstrated that sorption isotherms become more linear when an organic solvent is added to a soil, probably the result of elimination of micro-voids into which PAH can be incorporated, as was also found for an increase in temperature.

THERMAL TREATMENT VERSUS SOAKING

From Figure 3.2a it can be derived that, compared to soaking, thermal pretreatment led to a stronger increase in bioavailability in soil from Kralingen. In soil from Schijndel on the other hand, soaking increased the availability of 4-ring and 5-6 ring PAH, whereas thermal treatment had no effect. Possibly, thermal treatment enhances only degradation of contaminants sorbed into soil organic matter. This means that the bioavailability of PAH increases as a result of a transfer from slow sorption sites to fast sorption sites, either by redistribution of PAH or by transformation of the sorption sites themselves. Soaking seems to effect all PAH as a result of a large decrease in the partition coefficient, although the effects observed are much smaller than expected based on calculations.
The experiments show that biodegradation of hydrophobic organic contaminants can be enhanced by both thermal pretreatment and soaking. However, the increases in bioavailability in the soil samples tested are too small for application of thermal pretreatment and soaking in soil decontamination processes yet.

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4

Increased PAH Desorption by Thermal Pretreatment of Soil

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Biodegradation of hydrophobic organic contaminants in soil is often limited by slow desorption of these compounds from the soil. In this study, we tried to decrease the amount of PAH that shows slow desorption by subjecting the soil to a temperature increase (60 - 100 °C) for a short period of time (10 min. - 24 h) prior to desorption. After heating, desorption kinetics of several PAH were determined and fitted to a two-compartment model, consisting of a fast and slow desorption compartment. It was found that slow desorbing fractions had decreased after a thermal treatment. For example, the slow desorbing fraction of pyrene decreased from 80 % before thermal treatment to 58 % after a thermal treatment of 24 hr at 100 °C. Higher temperatures and longer treatment times led to smaller slow desorbing fractions. The results of this study correspond well with a mechanism in which contaminants that are entrapped within the soil organic matter are released as a result of a thermal pretreatment.

INTRODUCTION

Biological remediation of soils polluted with hydrophobic organic contaminants (HOC), like mineral oil and polycyclic aromatic hydrocarbons (PAH), often shows slow and incomplete degradation of these contaminants. This incomplete degradation is mainly caused by a slow desorption of the HOC from the soil and not by slow degradation itself [1,2].

It has often been shown that desorption of hydrophobic compounds from soil takes place in two stages: a first stage with fast desorption followed by a stage with slow desorption. The fraction of the contaminants that show fast desorption accounts for the part of the contaminants that can be removed by bioremediation, whereas the slow desorbing fraction accounts for the residual fraction in bioremediation [2]. In literature, this slow desorption has been explained by slow diffusion through rigid or glassy soil organic matter [3], slow desorption from micropores in the inorganic soil matrix [4,5], or entrapment of contaminant molecules either in nano-pores in the soil organic matter [6-8] or due to rearrangements of the soil organic matter structure [9,10].

In Chapter 2 and 3 several methods were investigated to increase the biodegradable part of the contaminants in order to improve bioremediation of PAH contaminated soils. It was found that a short thermal treatment of soil (1 h, 95-120 °C) increased the fraction of PAH which could be biodegraded. This increase of the biodegradable fraction was attributed to either a redistribution of sorbed contaminants from the slow desorption compartment to the fast one, or a transformation of the sorption compartments themselves in such a way that more contaminants exhibit fast desorption. In a previous study [11], no discrimination could be made between these causes for biodegradability increase after a thermal treatment.

In this study we tried to determine the effects of pretreatment time and temperature on contaminant desorption. Also we tried to further elucidate the mechanism that causes the increase of the PAH bioavailability as a result of thermal treatment. For this, desorption kinetics of PAH in thermally treated and non-thermally treated soils were studied. To determine desorption kinetics, a solid phase extraction method was used.

MATERIAL AND METHODS

SOILS

The desorption of PAH contaminants were studied in a sludge residue from a soil washing plant. The original soil had been obtained from a former gas plant site in Kralingen, the Netherlands. The soil had been separated at a cut-off diameter of 63 μm using hydrocyclones. The sludge residue - the product containing the smallest and lightest particles - was air dried and sieved at 2 mm to remove coarse, light material that had not been separated by hydrocyclonage. The total PAH concentration (16 US-EPA) was 115 mg/kg dry matter. The sludge also contained 300 mg mineral oil /kg dry matter and 9 mg cyanide/kg dry matter. The organic matter content of the sludge, determined by loss on ignition at 550 °C, was 12.3 %. The inorganic part of the sludge consisted of 27 % clay, 46 % silt and 27 % sand. The soil organic matter was expected to dominate PAH sorption for several reasons. First, the soil organic matter contents are relatively high compared to the oil concentrations, 13,000 mg/kg and 300 mg/kg, respectively. Consequently, the oil present in the soil most likely does not affect PAH desorption. Furthermore, the soil washing process, before sampling of the sludge, included a flotation step, which removed free organic material, like coal tar particles. Finally, sorption of PAH to mineral soil surfaces is unlikely for water contents above 0.1% [12]; the water content of the air-dried sludge was 3 %.

METHODS

Before the desorption experiments, 2 grams of air-dried sludge were brought in a serum flask that was closed with a Teflon lined screw cap. The serum flask was heated in a waterbath at a set temperature (60, 80, or 100 °C) during a set timespan (10, 30, 60 minutes, or 24 hours). After heating, the serumflask was cooled under running water.

After the thermal treatment the desorption kinetics of PAH from the sludge were determined in a sludge-water suspension. The method used for determining desorption kinetics was adopted from Cornelissen et al. [13] and Caroll et al. [3]. In this method a polymer adsorbent is used to extract desorbed compounds from the aqueous phase. In this way the contaminant concentration in the aqueous phase is kept very low and the maximal desorption rate will be achieved.

In this study, Tenax TA (Chrompack, mesh 20-35) was used as an adsorbent for PAH that had been desorbed from the soil. Because the density of Tenax is lower than that of water, it can easily be separated from the soil suspension. Before use

the Tenax was sieved at 200 μm to remove small particles, rinsed with acetone, and dried overnight at 30 °C.
For determining the desorption kinetics, the thermally treated sludge was transferred into a 50 ml separation funnel and 40 ml of 0.01 M $CaCl_2$ solution, 0.5 g of Tenax, and 10 mg NaN_3 (to prevent microbial degradation) were added. The separation funnel was shaken end-over-end (22 rpm, 30 °C) and at timed intervals (initially 1 hour, finally 3 days) the soil suspension was separated from the Tenax and fresh Tenax was added to the soil suspension. The residual Tenax was extracted with 20 ml of acetone on a horizontal shaker for 15 minutes (130 rpm). No more PAH could be extracted in a second extraction step, which means that all PAH could be extracted with a single-step extraction. PAH concentrations in the acetone were determined by HPLC. At the end of the experiment, residual PAH concentrations in the soil were determined by extracting the soil with 10 ml of 1-methyl-2-pyrrolidinone (99 %, Acros) in a microwave oven (MDS 2100, CEM corp.) at 130 °C for 1 hour. This extraction method was found to be faster and more efficient than traditional soxhlet extraction procedures [14]. After extraction the 1-methyl-2-pyrrolidinone was analysed by HPLC. The desorption experiments were carried out in duplicate.

ANALYTICAL EQUIPMENT

The PAH extracts were analysed using the following HPLC system: a GasTorr GT103 degassing device, a Gynkotech 480 HPLC pump, a Spark Holland Basic Marathon Autosampler, and a Gynkotech UVD340S photodiode array detector. A Vydac 5 C18 reverse phase column (250 mm × 4.6 mm L × ID) with external guard column and a solvent gradient program with acetonitrile (LabScan, HPLC-grade) and distilled water were used to separate PAH compounds. Concentrations were determined by UV absorbance at 254, 264, 287 and 335 nm. Peaks were integrated using Gynkotech Chromeleon 4.20.

DATA INTERPRETATION

The desorption kinetics were described with a two compartment model consisting of a slow desorption and a fast desorption compartment. Desorption from each compartment is assumed to be first order:

$$\frac{S_t}{S_0} = F_{slow} e^{-k_{slow} t} + F_{fast} e^{-k_{fast} t} \qquad (4.1)$$

where S_t and S_0 are the concentrations of soil sorbed PAH at time t and at the start of the experiment, respectively; F_{slow} and F_{fast} are the initial fractions of the contaminants in the slow and the fast desorbing compartment, respectively (notice: $F_{slow} = 1\text{-}F_{fast}$); k_{slow} and k_{fast} are the first-order rate constants of slow and fast desorption, respectively. Values for F_{slow}, F_{fast}, k_{slow} and k_{fast} were determined by calculating the least sum of squared differences between the calculated and experimental values of S_t/S_0.
The parameters of the two-compartment model will only give a unique description of the desorption curves in case $k_{fast} >> k_{slow}$. This condition was assured before discussion of the desorption results.

RESULTS AND DISCUSSION

DESORPTION CURVES

Typical desorption curves of pyrene (4-ring PAH compound) are given in Figure 4.1. This figure also shows the curve fitted with the two-compartment model. R^2 values for the fitted and observed desorption curves were 0.997 and 0.999 for the non-treated and the thermally treated soil, respectively. For all experiments and all PAH compounds, R^2 values ranged from 0.98 to 0.999, which shows that the two-compartment model can describe the data very well. As mentioned before, it is only allowed to use the two-compartment model for the description of desorption curves in case $k_{fast} >> k_{slow}$.

Figure 4.1

Desorption of pyrene from non-pretreated (□) and two thermally treated soil samples. Treatment times were 1 hour (○) and 24 hours (◇). The lines show the best fit with the two-compartment model.

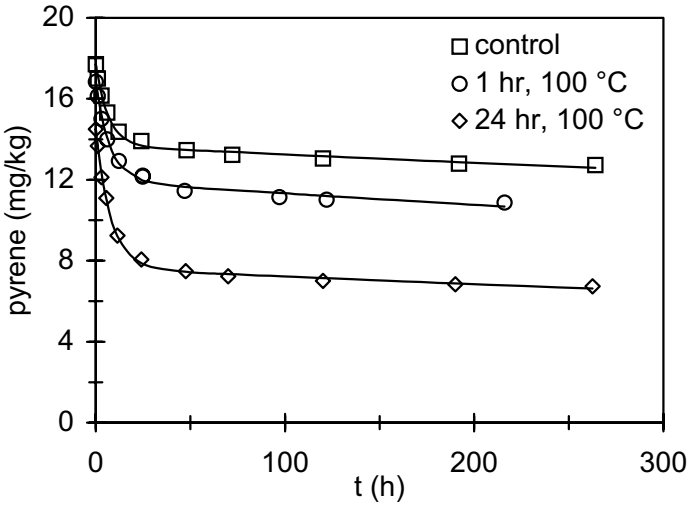


Table 4.1 shows that k_{fast} values are more than 100 times larger than k_{slow} values. Thus, it can be concluded that the two-compartment model can be used to describe desorption kinetics. Values in Table 4.1 are averaged for all desorption experiments, because the desorption rate constants for the different pretreatments were not significantly different (Student t-test, $p>0.1$) and no trend with increasing treatment temperature or increasing treatment time could be observed. Table 4.1 shows that both k_{fast} and k_{slow} decrease with increasing hydrophobicity of the PAH compounds (phenanthrene $\rightarrow$ chrysene). This decrease is more substantial for k_{fast} than for k_{slow}.

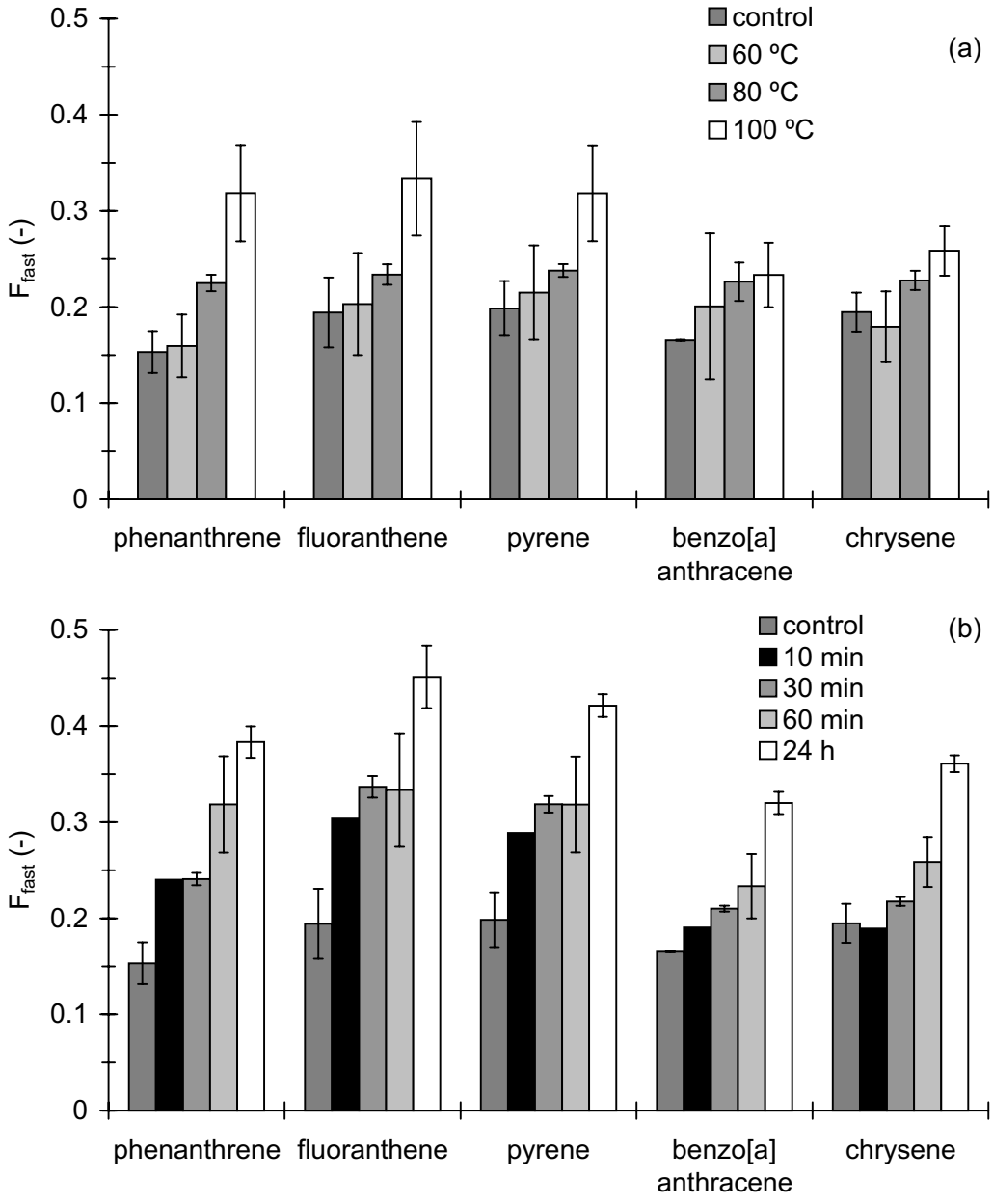
Values of F_{fast} for phenanthrene, fluoranthene, pyrene, benzo[a]anthracene, and chrysene are given in Figures 4.2a and b. Fast desorbing fractions are larger after a thermal treatment than before a thermal treatment (significant for 1 hr at 80 °C, 1 hr and 24 hr at 100 °C; Student t-test, $p<0.05$). These results are comparable to the increases of the biodegradable fractions after a thermal pretreatment, like has been shown in Chapter 3. It can further be seen that the increase of the fast desorbing fractions is dependent on the treatment temperature and treatment time. Figure 4.2a shows that higher temperatures lead to larger fast desorbing fractions. Similar results were found for the effects of treatment time; Figure 4.2b shows that longer times lead to larger fast desorbing fractions also (effects of temperature and time are significant for total PAH, not for individual compounds, Student t-test, $p<0.05$).

Table 4.1 Average values and standard deviations for k_{fast} and k_{slow} for all desorption experiments (n=12); diffusion coefficients, octanol-water partition coefficients, and organic carbon-water partition coefficients of the PAH studied.

PAH	k_{fast} (h^{-1})	k_{slow} (10^{-3} h^{-1})	D_b (10^{-9} m^2/s)	*log* K_{ow}	*log* K_{om}
phenanthrene	0.29 ± 0.07	0.51 ± 0.11	1.41	4.52	3.74
anthracene	0.20 ± 0.09	0.44 ± 0.19	1.42	4.50	4.73
fluoranthene	0.116 ± 0.024	0.41 ± 0.11	1.35	5.22	4.25
pyrene	0.098 ± 0.018	0.42 ± 0.12	1.34	5.12	4.17
benzo[a]anthracene	0.043 ± 0.006	0.38 ± 0.11	1.23	5.81	4.67
chrysene	0.043 ± 0.009	0.32 ± 0.15	1.23	5.71	4.60

Figure 4.2

Fast desorbing fractions of several PAH compounds for thermally treated and non-treated Kralingen soil.
(a) effects of treatment temperature (t = 60 min)
(b) effects of treatment time (T = 100 °C)
Error bars indicate 1 standard deviation. No error bars are shown for the control of benzo[a]-anthracene and the 10 minute thermal treatment at 100 °C; no duplicate sample.



To elucidate the nature of the increase of the fast desorbing fractions, it is necessary to discuss the mechanisms underlying the fast and slow desorption processes.

FAST DESORPTION

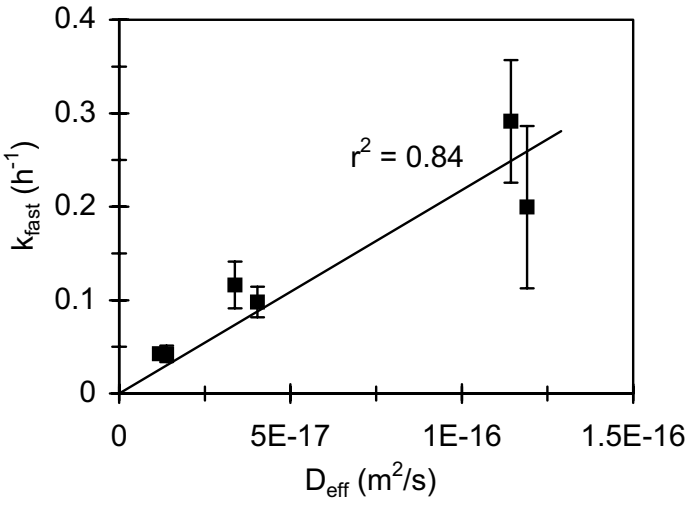
Table 4.1 showed that k_{fast} has a strong correlation with the hydrophobicity of the desorbing PAH compounds. This strong relation would be expected for a retarded pore diffusion process, in which diffusion of contaminants through soil aggregate pores is retarded by sorption to soil organic matter inside these pores [15]. The effective pore diffusion coefficient for retarded pore diffusion can be calculated using the following equation [16]:

$$D_{eff} = \frac{D_b \cdot \varepsilon}{(1-\varepsilon) \cdot \rho \cdot f_{oc} \cdot K_{oc}} \qquad (4.2)$$

where D_b is the diffusion coefficient of the organic compound in the aqueous phase (m^2/s); ε is the porosity of the soil aggregate (-); ρ is the density of the solid soil material (kg/m^3); f_{om} is the organic matter content of the soil (-); K_{om} organic matter water partition coefficient (m^3/kg).
Figure 4.3 shows k_{fast} against the effective diffusion coefficient for pore diffusion (D_{eff}). The relation between k_{fast} and D_{eff} as shown in this figure is nearly proportional ($R^2 = 0.84$), like would be expected for pore diffusion processes.
D_b in eq. 4.2 was calculated from the molecular volumes of the PAH compounds as mentioned in [17]. K_{om} was calculated from the octanol-water partition coefficient (K_{ow}) according to [18]. K_{ow} values were obtained from [19].

Figure 4.3

Fast desorption rate constants as a function the effective diffusion coefficient
Error bars indicate 1 standard deviation; they may fall within the symbol. Solid line shows best proportional fit.



The porosity of the particles was set on 0.12, in accordance with Wu and Gschwend [16] who found porosities from 0.07 to 0.17 for clay soils. The density of the soil material was set on $2.5 \cdot 10^3$ kg/m^3. Values of D_b, K_{ow}, and K_{om} for different PAH compounds are given in Table 4.1.
Assuming soil aggregates being spheres, the aggregate diameter can be calculated using k_{fast} and D_{eff} using an equation for radial diffusion [20]. For all PAH compounds the calculated aggregate diameters were found to be near 11 μm, which is the size of small clay aggregates. This realistic value of the calculated aggregate diameter strengthens the hypothesis that fast desorption is governed by retarded pore diffusion.

SLOW DESORPTION

Table 4.1 shows that values of k_{slow} for the different PAH compounds are far less dependent on K_{ow} values than k_{fast}. Similar results can be extracted from a paper by Cornelissen et al. [13], who studied not only PAH but also chlorobenzenes (CB) and polychlorobiphenyls (PCB). Their data on the desorption of these three groups of compounds (PAH, CB and PCB) span a wider range of values for both k_{slow} and K_{ow} than the data from this study and showed no correlation between k_{slow} and K_{oc}. This gives reason to believe that a different mechanism than aqueous diffusion causes slow desorption.
As mentioned before, proposed causes of slow desorption are slow diffusion through rigid or glassy soil organic matter [3], slow desorption from micropores in the inorganic soil matrix [4,5] or entrapment of contaminant molecules within the soil organic matter, either in nano-pores in glassy organic matter [6-8] or due to rearrangements of the soil organic matter structure [9,10]. For all three mechanisms, the mass transfer rate should be relatively independent on the hydrophobicity (K_{ow}) of the desorbing compound. The results of this study can not discriminate between the different mechanisms.

MECHANISM FOR INCREASE OF F_{FAST} AFTER THERMAL PRETREATMENT

The increase of F_{fast} after a thermal pretreatment corresponds with findings of Xing and Pignatello [7] who studied sorption isotherms of hydrophobic organics to soil organic matter. They found that these isotherms became more linear with increasing temperature. Several authors have shown that the non-linear parts of these isotherms are related to slow desorption processes [8,21]. Altogether, this means that the slow desorption compartment becomes smaller with increasing temperatures.

The decrease of the non-linear part of the isotherms and, consequently, the decrease of the slow desorption compartment was attributed to the removal of nano-sized pores in the soil organic matter, as a result of a glass-rubber transition of (part of) the soil organic matter. Entrapment of hydrophobic compounds inside the nano-pores is often suggested to cause slow desorption [6,8]. A glass-rubber transition of soil organic matter, similar to glass-rubber transitions in synthetic polymers, has previously been reported by LeBoeuf and Weber [22].

However, our results do not comply completely with this glass-rubber transition hypothesis for two reasons. First, a glass-rubber transition is a reversible process [23], which means that the decrease of the slow desorption fractions after temperature rise should be undone after returning to lower temperatures. Consequently, no effect of thermal treatment would be expected, which contrasts with the results from this study that showed that fast desorbing fractions had increased after a thermal treatment. Unfortunately, Xing and Pignatello had not determined desorption isotherms after the soil had returned to room temperature, so it could not be seen whether the linearisation of the sorption isotherms is a reversible process or not. Further, a glass-rubber transition is an instantaneous process [23], whereas the results presented here showed that a longer thermal treatment time leads to a larger increase of the fast desorbing fraction. Therefore, it is unlikely that a glass-rubber transition causes an increase of the fast desorbing fractions after a thermal treatment.

An alternative mechanism for slow desorption that might explain our results was suggested by Kan et al. [10,24] and Schlebaum et al. [9]. They stated that slow desorption is caused by the entrapment of organic compounds due to a structural rearrangement of the soil organic matter.

Examples of entrapment using 3D molecular modelling calculations are given by Kubicki and Apitz [25] and by Schulten [26]. They showed that napthalene and DDT, respectively, can become entrapped within structures of dissolved and soil bound organic matter. The driving force for this entrapment was the energy minimisation of the molecular interactions between the different organic groups of the soil organic matter and between the soil organic matter and the contaminant molecules. Kubicki and Apitz further showed that the entrapments were less strong at elevated temperatures. With a temperature increase, entropic contributions to the total energy of the organic matter-contaminant complex will become more important, which favours a less tight conformation of the organic matter. Consequently, contaminant molecules are no longer trapped within the soil organic matter. According to this theory, a thermal pretreatment will then lead to a decrease of the amount of entrapped contaminants and thus to a decrease of the slow desorbing fractions. Moreover, it can be expected that

longer treatment times and higher treatment temperatures lead to an increased release of entrapped contaminants, because the formation of a less tight conformation of the soil organic matter requires the movement of soil organic matter groups and is therefore a kinetically limited process.
Summarising, a thermal pretreatment of contaminated soil leads to larger fast desorbing fractions of organic contaminants, which is most probably caused by release of contaminants which were entrapped in the soil organic matter. Higher treatment temperatures and longer treatment times lead to larger fast desorbing fractions. Consequently, a larger fraction of PAH contaminations can be removed by biodegradation after a thermal treatment and more contaminated soils can be bioremediated to reach legal standards.

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5

Limitations for PAH Bioremediation in Soil and Sediment: Desorption versus Biodegradation

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A study was performed to determine whether desorption experiments (using solid-phase extraction) can predict bioremediation results. Both desorption and biodegradation experiments were performed with soil slurries, which made it possible to directly compare biodegradation and desorption kinetics. Two different soils and one sediment were used in the experiments, which all were field-contaminated. It was found that not only the fast desorbing PAH could be biodegraded, but also part of the slowly desorbing PAH. Further, biodegradation rates initially controlled removal of both low and high molecular weight PAH compounds. Nevertheless, eventually after 21 days, desorption rates controlled biodegradation of the low molecular weight PAH, and consequently residual concentrations of these compounds were the same for both desorption and biodegradation experiments. This showed that desorption experiments eventually predict biodegradation results for low molecular weight PAH well. On the other hand, for high molecular weight PAH compounds, biodegradation rates were always smaller than desorption rates, which was concluded to be caused by low growth rates of microorganisms that have to degrade these compounds.

submitted to *Bioremediation Journal*

INTRODUCTION

Bioremediation of soils contaminated with polycyclic aromatic hydrocarbons (PAH) has often been unsuccessful due to slow degradation rates and high residual contaminant concentrations that do not meet legal standards [1,2]. It has been suggested that bioremediation of PAH contaminated soils is limited by desorption of PAH from the soil and not by degradation of PAH by soil microorganisms [3-6]. The reason for this is that most PAH can be readily degraded when they are dissolved in water [7,8], whereas the presence of soil greatly reduces degradation rates of PAH [6,9,10].

Desorption of PAH and other hydrophobic contaminants from soil generally take place in two stages: first, a stage of fast desorption, followed by a phase of slow desorption [11,12]. For the fraction of the contaminants that shows fast desorption, bioremediation may be limited by the biodegradation rate, whereas for the slowly desorbing part, the desorption rate itself is limiting remediation. The fast desorbing part is said to account for the bioavailable part of the contaminants present [4,5].

Very few studies have directly compared desorption rates and degradation rates of soil sorbed PAH. Cornelissen et al. [4] compared PAH desorption in soil slurries to residual concentrations of PAH after biodegradation in bioreactors and biopiles. They found that desorption determines the extent of biodegradation for most PAH, except for 5-6 ring PAH, for which unspecified microbial factors were said to limit removal. Others have also suggested that removal of 5-6 ring PAH from soil might be limited by a lack of degradability of these compounds [7,8]. A study on biodegradation and desorption of α-hexachlorocyclohexane was performed by Rijnaarts et al. [13] who compared desorption and biodegradation rates and found that initially biodegradation limited removal and in a later stage desorption limited removal.

In this study, desorption and biodegradation rates of PAH are compared in order to determine which factors limit remediation of PAH compounds in soil. The experimental set-ups of desorption and biodegradation experiments in this study make it possible to compare desorption and biodegradation kinetics. It is investigated whether desorption experiments can be used to predict bioremediation results. In the case where biodegradation was found to limit remediation it is tried to elucidate which factors cause slow biodegradation.

MATERIAL AND METHODS

MATERIALS

Desorption and biodegradation of PAH contaminants were studied in three different soil or sediment samples. All three samples had been used in previous studies [14,15]. The first sample (referred to as KGP) was a sludge residue from a soil washing plant. The original soil had been obtained from a former gas plant site in Kralingen, the Netherlands. The soil had been separated at a cut-off diameter of 63 μm using hydrocyclones. The second sample (referred to as SWP) was a sandy soil from a wood preservation site in Schijndel, the Netherlands. The last sample (referred to as PHS) was a dredged sediment from the 1st Petroleum Harbour, Amsterdam. PAH and oil contaminations in this sediment originate from a large spill at the end of World War II. Organic matter contents and PAH concentrations of the different soils are shown in Table 5.1. KGP and SWP were air dried, crushed and sieved at 2 mm before use. PHS was not dried and crushed, but was only sieved at 2 mm.

BIODEGRADATION EXPERIMENTS

The biodegradation experiments were performed as described in more detail in Chapter 3. Briefly, 5 g (d.m.) of soil were added to 60 ml serumflasks together with 2.5 ml inoculum of an enrichment culture. The inoculum was grown on highly PAH-contaminated sediment for 15 days and it showed to be capable of degrading all 16 EPA-PAH compounds, except indeno[1,2,3-cd]pyrene. The total liquid volume was adjusted to 10 ml with a mineral medium solution. The inoculated samples were incubated at 30 °C and mixed on an end-over-end shaker at 22 rpm. Regularly, the oxygen content was determined by GC to verify it was not limiting biodegradation. If necessary the headspaces were flushed with compressed air or pure oxygen. At timed intervals, flasks were removed from the shaker and sacrificed for analysis. PAH were extracted from the soil suspension with 40 ml acetone at room temperature during 1 hour [16]. The extracts were analysed for PAH by HPLC. Biodegradation experiments were carried out in triplicate (KGP and SWP) or quadruplicate (PHS).

DESORPTION EXPERIMENTS

The method for determining desorption kinetics of PAH from soil was adapted from Cornelissen et al. [12] and Caroll et al. [17] and has been described in more detail in Chapter 4. In this method a polymer adsorbent (Tenax) is used to extract desorbed PAH from the water phase. In this way the contaminant concentration in the aqueous phase is kept very low, and a maximum concentration gradient between the soil and the aqueous phase is maintained. This ensures that maximum desorption rates are achieved [12].

Briefly, 2 g (d.m.) of soil were put in a separating funnel together with 40 ml of a 0.01 M $CaCl_2$ solution, 0.5 g of Tenax, and 10 mg of NaN_3. The funnel was mixed end-over-end at 30 °C and at timed intervals the soil suspension was separated from the Tenax and 0.5 g of fresh Tenax was added to the soil suspension. PAH were extracted from the contaminated Tenax with 20 ml of acetone in a single extraction step. Subsequent extraction steps yielded no more PAH. PAH concentrations in the acetone were determined by HPLC. At the end of the desorption experiments the soil suspension was centrifuged and the residual PAH concentrations in the soil were determined after extraction from the soil with 10 ml of 1-methyl-2-pyrrolidinone (NMP) in a microwave oven at 130 °C during 1 hour; extraction efficiencies with NMP are comparable to efficiencies using acetone extraction [16].

Table 5.1 Organic matter contents and PAH contents of soils used in this study

soil sample	*organic matter*[a] *(%)*	*PAH (16-EPA) (mg/kg)*	*2-3 ring PAH (mg/kg)*	*4 ring PAH (mg/kg)*	*5-6 ring PAH (mg/kg)*
KGP	12.3	121	46	49	26
SWP	1.0	101	63	54	6
PHS	9.9	2088	1421	519	147

[a] in percentage of dry weight determined after loss on ignition at 500°C

NMP, instead of acetone, was used for extraction, because the water content of the soil before extraction was unknown; the water content was found to be an important parameter for acetone extraction [16]. The concentrations of all 16 EPA-PAH in the extracts were determined by HPLC. Desorption experiments were carried out in duplicate.

DATA INTERPRETATION

Desorption of PAH from soil generally follows a so-called biphase pattern: first, a phase of fast desorption, followed by a phase of slow desorption [11,12]. A two-compartment model consisting of a slowly desorbing and a fast desorbing compartment was used to describe these biphase desorption kinetics. Desorption from each compartment was assumed to be first order:

$$\frac{S_t}{S_0} = F_{slow}\, e^{-k_{slow}t} + F_{fast}\, e^{-k_{fast}t} \qquad (5.1)$$

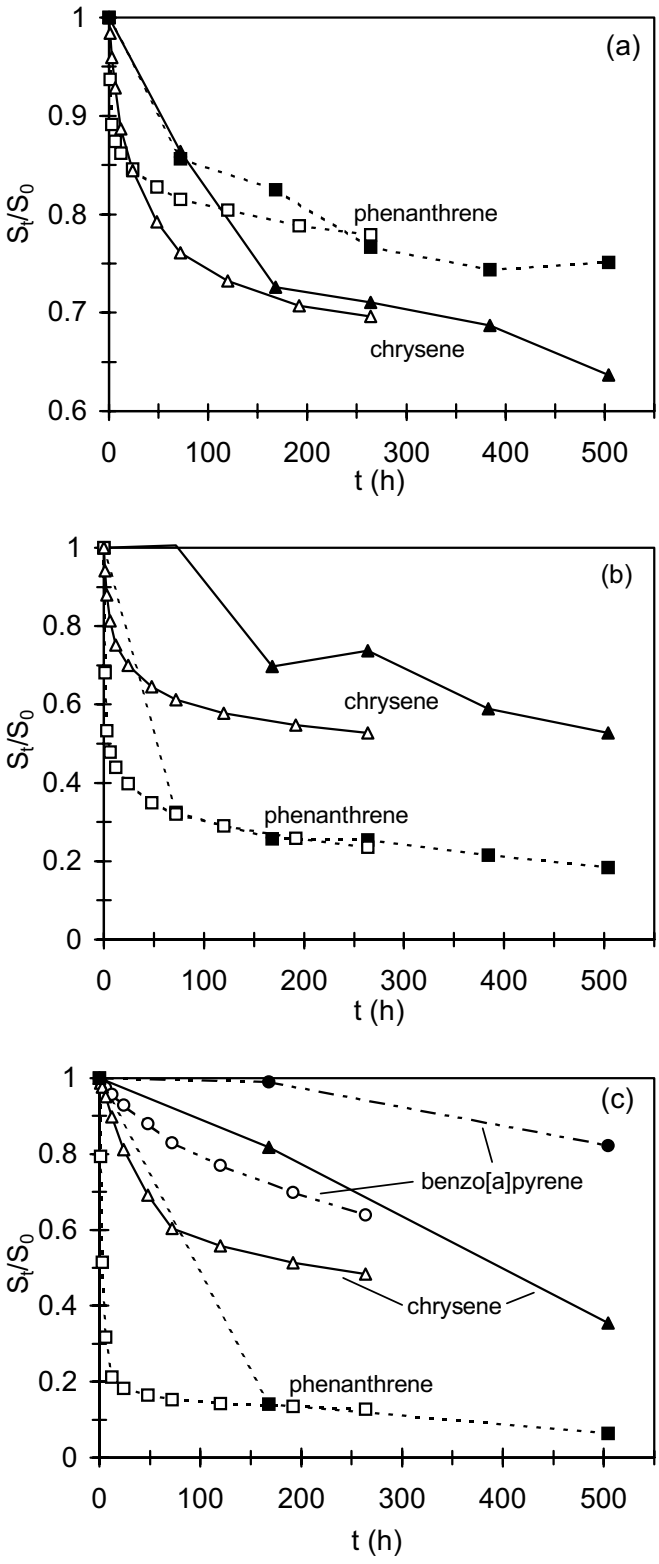
where S_t and S_0 are the concentrations of PAH in the soil at time t and at the start of the experiment, respectively (mg/kg); F_{slow} and F_{fast} are the initial fractions of the contaminants in the slowly and the fast desorbing compartment (notice: $F_{slow} = 1\text{-}F_{fast}$); k_{slow} and k_{fast} are the first-order rate constants of slow and fast desorption (h^{-1}). Values for F_{slow}, F_{fast}, k_{slow} and k_{fast} were determined by calculating the least sum of squared differences between the calculated and experimental values of S_t/S_0.

RESULTS

Degradation and desorption curves of selected PAH in KGP, SWP, and PHS samples are shown in Figures 5.1a, b, and c, respectively. Phenanthrene (3-ring PAH) and chrysene (4-ring PAH) could be detected in all three soils and are shown in all three figures. Phenanthrene was the smallest PAH compound and chrysene the largest PAH compound that could be detected in KGP and SWP. In PHS higher molecular weight PAH could be detected also. Figure 5.1c shows the degradation and desorption curves of benzo[a]pyrene (5-ring PAH) for PHS.
These figures show that all three PAH compounds exhibited an initial phase of fast desorption followed by a phase of slow desorption. This biphase desorption behaviour was described very well with eq. 5.1: values for R^2 between the fit and the experimental data of all PAH ranged from 0.980 to 0.999 for all experiments.

Figure 5.1

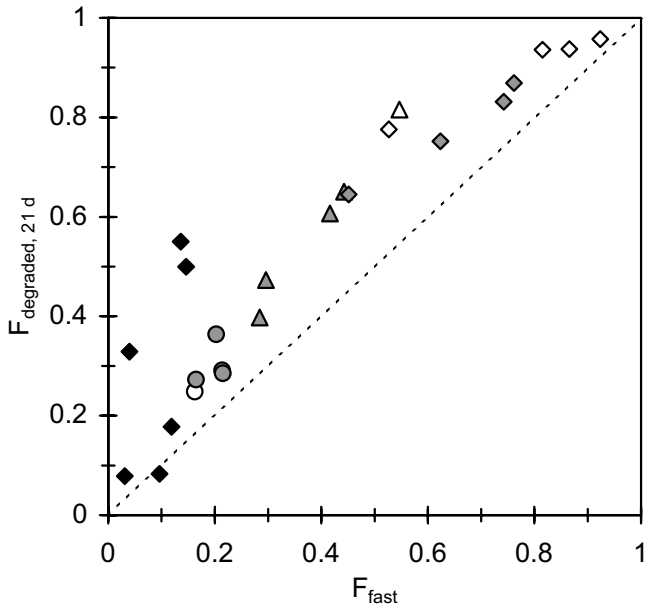
Desorption (open symbols) and degradation (closed symbols) of phenanthrene (squares), chrysene (triangles) and benzo[a]pyrene (circles) in three different soil samples:
(a) KGP
(b) SWP
(c) PHS



Comparable biphase patterns were found for the biodegradation curves, although, the patterns were not always as pronounced as in the desorption experiments.
Both the initial desorption rates and the initial degradation rates were higher for phenanthrene than for chrysene in all soils. In PHS, benzo[a]pyrene showed the lowest desorption and degradation rates. In KGP more chrysene than phenanthrene was desorbed and degraded at the end of the experiments. The opposite was found in SWP and PHS. It is speculated that some desorption or degradation of the more soluble phenanthrene had taken place in KGP prior to sampling of the soils.
For phenanthrene, initial degradation rates and initial desorption rates were comparable (Student t-test, 1-sided, 95 %, at t = 72 hours). For both chrysene and benzo[a]pyrene, initial biodegradation rates were lower than the desorption rates (Student t-test, 1-sided, 95 %). However, final degradation and desorption rates of chrysene were comparable. The biological removal of benzo[a]pyrene was much slower than desorption and also final concentrations in the biodegradation experiment were much higher than in the desorption experiment. This suggests that for low and medium molecular weight PAH, like phenanthrene and chrysene, biodegradation determined removal rates only initially, whereas at the end of the biodegradation experiment, the desorption limited removal. In contrast to this, the removal rate for high molecular weight compounds, like benzo[a]pyrene, was always limited by the biodegradation rate.

Figure 5.2

Fractions of PAH compounds that show fast desorption (F_{fast}) and fractions degraded in 21 days ($F_{degraded,21d}$); three different soils: KGP (circles), SWP (triangles) and PHS (diamonds); PAH are divided in 3 groups: 2-3 ring (white), 4 ring (grey) and 5-6 ring (black) compounds



DISCUSSION

PREDICTION OF BIODEGRADATION USING DESORPTION EXPERIMENTS

As mentioned, the biphase desorption patterns in Figures 5.1a-c was described very well with eq. 5.1. Values of the fast desorbing fractions (F_{fast}) were determined for all PAH by fitting eq. 5.1 to the observed desorption curves. Several authors have suggested that the fast desorbing fractions account for the biodegradable part of the contaminants [4,5]. To verify this, F_{fast} was plotted against the fractions that had been biologically degraded after 21 days ($F_{degraded,21d}$) in Figure 5.2.

Assuming that the fast desorbing fraction is a good predictor of the degraded fraction, all data points should be close to the dotted line. Figure 5.2 shows that all but one of the points are above this line, which means that for almost all PAH compounds in all three soils $F_{degraded,21d}$ is larger than F_{fast}. This indicates that not only the fast desorbing fractions of the contaminants were degraded but also part of the slowly desorbing fractions. Therefore, a better predictor of the biodegradation results might be the total fraction of the contaminants that had desorbed after 21 days ($F_{desorbed,21d}$). These fractions were calculated by extrapolating the desorption curves using eq. 5.1 and are compared with $F_{degraded,21d}$ in Figure 5.3. This figure shows that $F_{desorbed,21d}$ predicts biodegradation results much better than F_{fast} , especially for 2-3 ring and 4 ring PAH. A t-test (2-sided, $p>0.01$) confirmed that the total desorbed fractions ($F_{desorbed,21d}$) are a better predictor of the biodegradable fractions ($F_{degraded,21d}$) than the fast desorbing fractions (F_{fast}). The test showed that F_{fast} and $F_{degraded,21d}$ were significantly different, whereas $F_{desorbed,21d}$ was not significantly different from $F_{degraded,21d}$.

Figure 5.3 further shows that data points of some 5-6 ring PAH are below the dotted line, which means that the fractions that were biodegraded are overestimated by the fractions that desorbed in 21 days. This means that for these compounds the removal rate was limited by the biodegradation rate.

Analogous to Figure 5.3, graphs of biodegradation versus desorption can be constructed for other periods of biodegradation and desorption. Figure 5.4 shows the fractions which were degraded after 7 days ($F_{degraded,7d}$) versus the fractions which had desorbed after 7 days ($F_{desorbed,7d}$). From this figure it can be seen that for most 2-3 ring PAH compounds $F_{desorbed,7d}$ gives a good prediction of $F_{degraded,7d}$. On the other hand, for the 5-6 ring PAH compounds and also for the 4 ring PAH, the fractions that had desorbed were larger than the fractions that had been biodegraded. Moreover, hardly any biodegradation of the 5-6 ring

compounds could be detected after 7 days. From these results can be concluded that removal of 2-3 ring and 4 ring PAH is mainly limited by desorption from soil, after a short period of removal limited by biodegradation. In contrast to this, for 5-6 ring PAH biodegradation seems to determine removal rates during the complete biodegradation experiments (Figures 5.1c, 5.3 and 5.4).

Figure 5.3

Fractions of PAH compounds that have been desorbed ($F_{desorbed,21d}$) and degraded ($F_{degraded,21d}$) in 21 days; three different soils: KGP (circles), SWP (triangles) and PHS (diamonds); PAH are divided in 3 groups: 2-3 ring (white), 4 ring (grey) and 5-6 ring (black) compounds

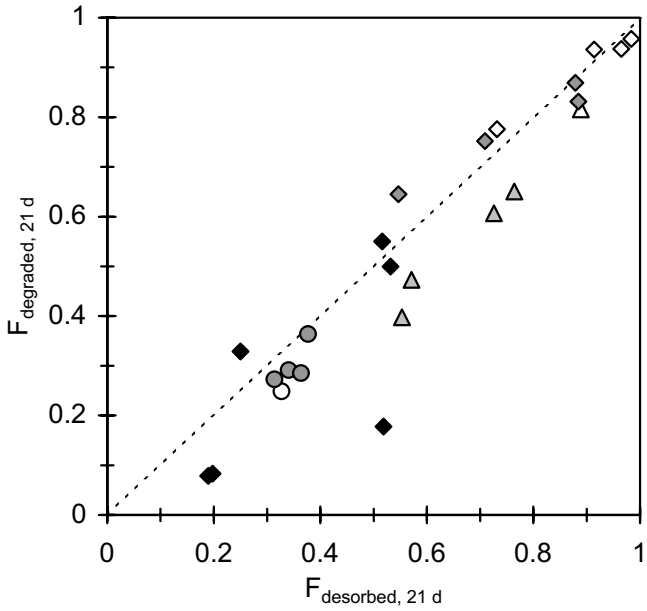
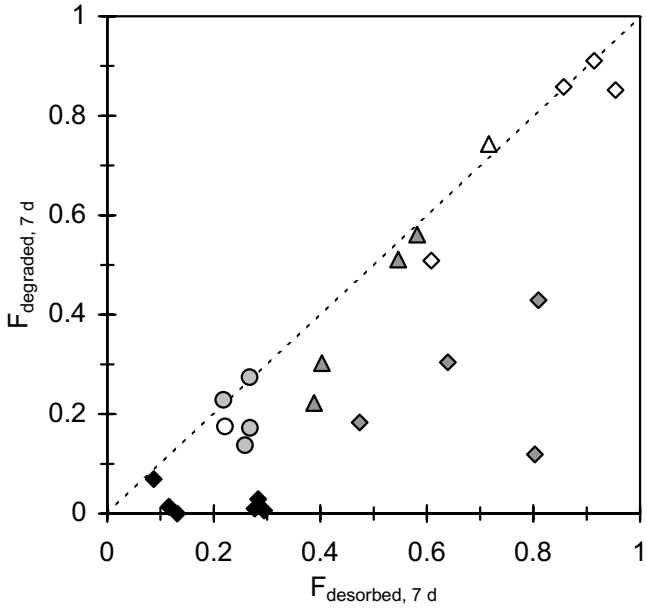


Figure 5.4

Fractions of PAH compounds that have been desorbed ($F_{desorbed,7d}$) and degraded ($F_{degraded,7d}$) in 7 days; three different soils: KGP (circles), SWP (triangles) and PHS (diamonds); PAH are divided in 3 groups: 2-3 ring (white), 4 ring (grey) and 5-6 ring (black) compounds



Others have also found that hardly any degradation of 5-6 ring PAH compounds takes place in soil systems [7,8]. There are several reasons which can cause this slow degradation: (a) a positive or slightly negative Gibbs free energy of the degradation, (b) a high activation energy for degradation, (c) the free concentrations of the compounds in the aqueous phase are too low for growth of micro-organisms. For 5-6 ring PAH, these three reasons will be discussed below on their probabilities to limit biodegradation rates.

McFarland and Sims [18] showed that the free energy of oxidation is only slightly different for different PAH compounds. The free energies of oxidation are around -105 kJ/electron equivalent for all PAH compounds. This means that the differences in degradation rates for different PAH can not be explained by differences in free energy.

Secondly, the activation energy for oxidation is determined by the ionisation potential of the PAH compounds. Hammel et al. [19] and Vazquez-Duhalt et al. [20] showed that a high ionisation potential prevents fungal degradation of some PAH compounds. However, the highest ionisation potentials are found for low molecular weight PAH [20,21], which means that the activation energy for oxidation of low molecular weight PAH is higher than the activation energy for oxidation of high molecular weight PAH. The degradation of high molecular weight PAH therefore is unlikely to be limited by this activation energy.

Finally, it has been shown that at very low substrate concentrations very slow growth or even no growth of the microbial population will occur [22,23]. This is due to the fact that part of the energy liberated from the degradation of substrate will not be used for microbial growth, but is used by the micro-organisms for maintaining basic microbial processes (osmotic regulation, regeneration of proteins and nucleic acids, molecular transport, etc.). At very low substrate concentrations, all energy from substrate degradation is needed for maintenance and no growth of the microbial population will occur. This means a threshold concentration for microbial growth can be determined [23-25]. For PAH degradation in soil, substrate concentrations are the free aqueous concentrations of the PAH. Even for micro-organisms attached to solid PAH surfaces, dissolving of the PAH precedes uptake by the micro-organisms [26]. Aqueous concentrations of 5-6 ring PAH are much lower than the concentrations of low molecular weight PAH, due to lower solubilities and higher soil-water partition coefficients of the 5-6 ring PAH [27]. Consequently, growth rates of micro-organisms degrading 5-6 ring PAH as primary substrate will most likely be lower than growth rates of degraders of low molecular weight PAH. These low growth rates will then lead to small or decaying microbial populations and consequently

to biodegradation rates lower than maximal desorption rates, as has been found in this study. To a lesser extent this can also be applied to biodegradation of 2-3 ring and 4 ring PAH, as in this study was found that biodegradation also limits removal of these PAH initially (Figures 5.1a-c, 5.4).

Summarising, desorption experiments can be used to predict bioremediation results for 2-3 ring and 4 ring PAH, because desorption rates eventually determine removal rates of these compounds from soil. For high molecular weight PAH compounds, biodegradation rates were always smaller than desorption rates, probably due to low growth rates of micro-organisms which had to degrade these compounds.

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6

Modelling PAH Removal in Soils and Sediments: Coupled Biodegradation and Desorption

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A mathematical model was constructed to describe desorption and biodegradation of polycyclic aromatic hydrocarbons (PAH) and growth of PAH-degrading biomass. Desorption of PAH was described by biphase desorption kinetics incorporating a fast and a slow desorption compartment, as is generally found in PAH desorption. Biodegradation and biomass growth were described using Monod kinetics to which a term for maintenance requirements was added. Modelling results were compared to experimental results of PAH biodegradation and desorption in three different soils. It was found that the model could describe the experimental biodegradation results well. Especially, the model showed that persistence of high molecular weight PAH in soil can be caused by slow growth of biomass degrading these substances. This slow growth was attributed to the low aqueous phase concentrations of these high molecular weight PAH, which were too low to meet the maintenance requirements of the micro-organisms. Further, the sensitivity of the model was tested for various input parameters. The model was found to be the most sensitive to the soil-water partition coefficients of the PAH and the maintenance or decay coefficients of the PAH degrading microorganisms.

INTRODUCTION

Polycyclic aromatic hydrocarbons (PAH) sorbed to soil are known to be very resistant to bioremediation [1,2]. The concentrations of PAH in the water phase are very low, due to sorption of these compounds to soil. Consequently, biodegradation rates are very low, as bacteria can only degrade dissolved compounds [3-5]. Biodegradation of soil sorbed PAH is further hampered by the low desorption rates [6,7]. In some cases biodegradation rates are lower than desorption rates, especially for high molecular weight PAH [7]. In several studies it has been hypothesised that these very low degradation rates are probably caused by the low aqueous phase concentrations of these high molecular weight PAH; these aqueous phase concentrations are too low to meet the maintenance requirements of the microorganisms [8-10]. As a result, no growth of the microbial population and consequently very slow degradation will occur.

To understand bioremediation results, it is necessary to determine under which conditions desorption governs remediation and when biodegradation does. Therefore, a model was developed to describe the desorption and biodegradation processes during bioremediation.

In several studies it has been tried to describe coupled sorption and biodegradation of hydrophobic organic compounds, often using first-order kinetics [8,10] or radial diffusion models [11,12] for desorption. However, for aged contaminations a fraction of the sorbed contaminants generally shows to be resistant for desorption. Desorption of this resistant fraction can be far better described using a biphase sorption model, which distinguishes a fast desorbing fraction and a slowly desorbing fraction of the contaminants (e.g. [13,14]).

Further, most studies on biodegradation of sorbed PAH only focussed on low molecular weight PAH compounds as naphthalene and phenanthrene. Biodegradation results of low molecular weight PAH could be well described using desorption kinetics, whereas biodegradation results of high molecular weight PAH could not [7]. Therefore, results of degradation experiments with low molecular weight PAH can not simply be extrapolated to degradation of high molecular weight PAH.

In this paper we present a model that describes coupled desorption and biodegradation of both low and high molecular weight PAH. Desorption kinetics of PAH are described with a biphase model. In the biodegradation kinetics, growth of PAH degrading biomass and maintenance requirements of the biomass are incorporated. Eventually, modelling results are compared to results from biodegradation and desorption experiments of several PAH contaminated soils.

MODEL

DESORPTION

Desorption of hydrophobic compounds from soil can be described very well with a biphase sorption model, in which a distinction is made between a fast and a slowly desorbing contaminant fraction. In this biphase model, desorption is described with two first order equations, one for each fraction, with different desorption rate constants for each fraction. In case the aqueous phase is kept free of desorbed compounds, like in many desorption experiments [13,15,16], desorption can be described with the following differential equations:

$$\frac{dS_{slow}}{dt} = -k_{slow} \cdot S_{slow} \tag{6.1}$$

$$\frac{dS_{fast}}{dt} = -k_{fast} \cdot S_{fast} \tag{6.2}$$

where, S_{slow} is the concentration of the slowly desorbing contaminants (mg/kg dry solids); S_{fast} is the concentration of the fast desorbing contaminants (mg/kg dry solids); k_{slow} is the slow desorption rate constant (h^{-1}); k_{fast} is the fast desorption rate constant (h^{-1}).
However, during biodegradation experiments the aqueous phase is not necessarily free of desorbed compounds. Then, desorption rates are determined both by the sorbed concentrations (S_{slow} and S_{fast}) and by the concentrations in the aqueous phase (C_w). Two desorption systems can be distinguished: a system in which desorption from the fast compartment is parallel to desorption from the slow compartment [17]:

$$S_{slow} \rightarrow C_w \leftarrow S_{fast} \tag{6.3}$$

or a system in which fast desorption is serial to slow desorption [18]:

$$S_{slow} \rightarrow S_{fast} \rightarrow C_w \tag{6.4}$$

Calculations showed that overall desorption rates were not much different for both systems (data not shown); in this study a serial desorption system was chosen, because parameters could be easier estimated. Desorption can now be described with the following differential equations:

$$\frac{dS_{slow}}{dt} = -k_{slow}\left(S_{slow} - K_{s\leftrightarrow f} \cdot S_{fast}\right) \quad (6.5)$$

$$\frac{dS_{fast}}{dt} = -k_{fast}\left(S_{fast} - K_p \cdot C_w\right) + k_{slow}\left(S_{slow} - K_{s\leftrightarrow f} \cdot S_{fast}\right) \quad (6.6)$$

$$\frac{dC_w}{dt} = r_{s,w} \cdot k_{fast}\left(S_{fast} - K_p \cdot C_w\right) \quad (6.7)$$

where, $K_{s\leftrightarrow f}$ is the equilibrium constant between the fast and the slowly desorbing compartment (-); C_w is the concentration in the aqueous phase (mg/l); K_p is the soil-water partition coefficient (l/kg); $r_{s,w}$ is the soil-water ratio (kg/l).

BIODEGRADATION

It is generally assumed that only compounds that are dissolved in the aqueous phase can be degraded by micro-organisms. Degradation of dissolved compounds can be described with Monod kinetics:

$$\frac{dC_w}{dt} = -\frac{B}{Y} \cdot \frac{\mu_{max} \cdot C_w}{K_s + C_w} \quad (6.8)$$

$$\frac{dB}{dt} = B \cdot \frac{\mu_{max} \cdot C_w}{K_s + C_w} - k_d \cdot B \quad (6.9)$$

where, B is the biomass concentration (mg/l); Y is the microbial yield (mg biomass/mg substrate); μ_{max} is the maximal growth rate of the biomass (h^{-1}); K_s is the affinity constant (mg/l); k_d is the biomass decay rate (h^{-1}). The k_d value is related to the maintenance coefficient, m (mg substrate/mg biomass·h) [8], which is often found in literature. The maintenance coefficient is the flow of substrate needed by the micro-organisms to maintain basic microbial processes (osmotic regulation, regeneration of proteins and nucleic acids, molecular transport, etc.):

$$k_d = m \cdot Y \quad (6.10)$$

Related to this is the threshold concentration ($C_{w,min}$ (mg/l)), below which no growth of the microbial population will occur [8,9]:

$$C_{w,min} = \frac{k_d \cdot K_s}{\mu_{max} - k_d} = \frac{m \cdot Y \cdot K_s}{\mu_{max} - k_d} \quad (6.11)$$

In case $C_w << K_s$ also first order kinetics can be used to describe biodegradation kinetics:

$$\frac{dC_w}{dt} = -\frac{B}{Y} \cdot k_{bio} \cdot C_w \qquad (6.12)$$

$$\frac{dB}{dt} = B \cdot k_{bio} \cdot C_w - k_d \cdot B \qquad (6.13)$$

where, k_{bio} (= μ_{max}/K_s) is the first order rate constant ($l \cdot mg^{-1} \cdot h^{-1}$).
For the model in this study, it has been assumed that a single bacterial species can only grow on a single PAH compound. This assumption is supported by work of Rogoff [19], who showed that for the first oxidation step of different PAH compounds different enzymes are required, although these enzymes could co-oxidise other PAH to a lesser extent. Similar results were reported by Bastiaens [20], who found that bacterial cultures grown on a specific PAH compound were not able to grow on other PAH compounds, but that each culture could cometabolise a few other PAH compounds to some extent. Cometabolism is not included in this model but its effects on the biodegradation results will be discussed further on. Combination of the differential equations for desorption (eqs. 6.5-6.7) and for biodegradation (eqs. 6.8 and 6.9, or eqs. 6.12 and 6.13) yields a model that can describe the desorption and subsequent biodegradation of hydrophobic, soil sorbed contaminants in a soil suspension. The model was numerically solved using a 4th order Runge-Kutta integration procedure.

PARAMETERS AND INITIAL VARIABLES

Modelling calculations have been performed for three different PAH compounds (phenanthrene, chrysene, and benzo[a]pyrene) in three different soils. These soils have been previously used in desorption and biodegradation experiments [21,22]. Initial PAH concentrations and soil characteristics are given in Table 6.1.
The desorption rate constants (k_{slow} and k_{fast}) were determined by fitting results from previous desorption experiments to eqs. 6.1 and 6.2. The desorption experiments have been extensively described in Chapter 4 and 5. The partition coefficients between the slowly and fast desorbing fractions ($K_{s\leftrightarrow f}$) were calculated from the initial fractions of the PAH that showed fast desorption (F_{fast}) and slow desorption (F_{slow}):

$$K_{s\leftrightarrow f} = \frac{F_{slow}}{F_{fast}} \qquad (6.14)$$

Experimental determined values for several PAH in the three soils of k_{slow}, k_{fast}, and F_{fast} are given in Table 6.2.
The soil-water partition coefficient (K_p) was calculated from the organic matter-water partition coefficient (K_{om}) and the soil organic matter content (f_{om}):

$$K_p = f_{om} \cdot K_{om} \qquad (6.15)$$

K_{om} was derived from the octanol-water partition coefficient using an empirical relationship determined by Karickhoff [23]. Calculated values for K_{om} are given in Table 6.2.
Values of biodegradation parameters are given in Table 6.3. Biodegradation kinetics had not been determined experimentally and therefore literature values had to be used for the model calculations. Only a few studies were found describing microbial degradation kinetics of PAH [24-27]. It was found that μ_{max} and K_s values were different for different PAH compounds, yet first order rate constants (μ_{max}/K_s) were similar [24,25,27]. Therefore, the same first order rate constants were used for different PAH compounds. It was legitimate to use first order biodegradation kinetics instead of Monod-kinetics, since C_w was always much lower than K_s (data not shown).
Values of biodegradation parameters are given in Table 6.3. Biodegradation kinetics had not been determined experimentally and therefore literature values had to be used for the model calculations. Only a few studies were found which describe microbial degradation kinetics of PAH [24-27].

Table 6.1 Characteristics of soils used in modelling calculations

name	*soil type*	*contamination source*	*org. matter*[a] *(%)*	*PAH (16-EPA) (mg/kg)*
KGP	clay	gas plant site	12.3	115
SWP	sandy	wood treatment plant	1.0	101
PHS	dredged sediment	oil spill	9.9	1940

[a] in percentage of dry weight determined after loss on ignition at 500 °C

It was found that μ_{max} and K_s values were different for different PAH compounds, yet first order rate constants (μ_{max}/K_s) were similar [24,25,27]. Therefore, the same first order rate constants were used for different PAH compounds. It was legitimate to use first order biodegradation kinetics instead of Monod-kinetics, since C_w was always much lower than K_s (data not shown).

Theoretical values for the microbial yield were calculated by McFarland and Sims [28]. They found a value of approximately 1 (mg/mg), which is comparable to the experimental values of 1.2 and 0.93 (mg/mg) found by Volkering [27] and Mulder et al. [26], respectively.

Values of k_d were calculated from the maintenance coefficients (m), using eq. 6.10. In literature a wide range of maintenance coefficients can be found: from 75 to 1500 ($J \cdot g^{-1}$ biomass$\cdot h^{-1}$) [27,29]. This is equivalent to m values of 0.002 to 0.04 (mg substrate$\cdot mg^{-1}$ biomass$\cdot h^{-1}$) [28], and therefore to k_d values of 0.002 to 0.04 (h^{-1}) using equation 6.10. It can be expected that maintenance coefficients of PAH degrading bacteria are relatively low, because these bacteria are likely adapted to low substrate concentrations and consequently are very efficient in their metabolism [8].

Finally, the initial biomass concentration could not be determined from experiments, and therefore this parameter was used to fit the model calculations to the results of biodegradation experiments.

Table 6.2 Parameters determining desorption kinetics

soil	*compound*	S_0 *(mg/kg)*	k_{fast} *(h^{-1})*	k_{slow} *(10^{-3} h^{-1})*	F_{fast}	*log* K_{om}
KGP	Phe	8.43	0.39	0.43	0.16	3.78
	Chr	7.94	0.053	0.48	0.21	4.95
SWP	Phe	6.52	0.72	2.8	0.55	3.78
	Chr	4.00	0.13	0.98	0.30	4.95
PHS	Phe	482	0.30	1.5	0.82	3.78
	Chr	55.3	0.022	0.38	0.45	4.95
	B[a]P	62.1	0.022	1.2	0.12	5.38

The parameters that determine the biodegradation kinetics (k_{bio} and k_d) and the organic matter-water partition coefficient (K_{om}) were not determined experimentally, but had to be derived from literature values. As a result of this, they are not as accurate as the parameters that determine desorption kinetics in this study. Therefore, calculations were carried out to test the sensitivity of the modelling results for these parameters. Biodegradation curves were calculated for values four times and one fourth of the mean values found in literature. It should be noted that the range of values used for the sensitivity analysis of k_{bio} is much larger than the range of values found in literature (see also Table 6.3), which makes the model less sensitive to variations in k_{bio} than will be suggested by the sensitivity analysis. Also calculations with different initial biomass concentrations were performed, because the initial biomass concentration could not be determined from experiments too.

RESULTS

Relative concentrations of phenanthrene, chrysene, and benzo[a]pyrene from modelling calculations and from previous desorption (Chapter 4 and 5) and biodegradation experiments (Chapter 3 and 5) are shown in Figure 6.1. Figures 6.1a-c show that experimental biodegradation results for phenanthrene follow desorption curves close for all three soils. Modelling calculations could describe this behaviour well for SWP and PHS. For KGP, the model predicted a slower biodegradation than was found in the experiments.

Table 6.3

Parameters determining biodegradation kinetics

parameter		*value*[a]	
k_{bio}	($l \cdot mg^{-1} \cdot h^{-1}$)	20	(10-40)
k_d	(h^{-1})	0.004	(0.002-0.04)
Y	(mg/mg)	1	(0.93-1.2)
B_0	(mg/l)	0.001	

[a]range of values found in literature are between brackets

Desorption and biodegradation results of chrysene are shown in Figures 6.1d-f. For all three soils, biodegradation was initially slower than desorption, but at the end of the experiments, biodegradation and desorption rates seemed similar. The modelling calculations for chrysene in SWP and PHS also showed a slow initial biodegradation, comparable to the experimental results. For KGP, the model predicted almost no biodegradation of chrysene, in contrast to the experimental results.

Benzo[a]pyrene could be only detected in PHS. Both the modelled biodegradation and the experimental biodegradation were slower than the experimental desorption. However, the model predicted almost no biodegradation whereas the experiments showed that 20 % of the benzo[a]pyrene was degraded within 500 hours.

Results of the sensitivity analysis for degradation of chrysene in PHS are shown in Figure 6.2. These figures show that the partition coefficient (K_{om}) and the first order rate constant for biomass growth (k_{bio}) have the largest effect on the biodegradation curves; larger values for K_{om} and smaller values of k_{bio} lead to slower degradation and higher final concentrations. The decay constant (k_d) has a smaller effect on the biodegradation rates and biodegradation endpoints. The initial biomass concentration has only a small effect on the period for which the biodegradation rate limits removal, and no effect on the final concentrations.

DISCUSSION

DEGRADATION RESULTS OF DIFFERENT SOILS

The results showed that the model described the experimental biodegradation of phenanthrene and chrysene in SWP and PHS well. On the other hand, for KGP the model predicted a slower biodegradation of both phenanthrene and chrysene than that was found in the biodegradation experiments. The reason for this might be a lower value of K_{om} for PAH in KGP, than in SWP and PHS, since K_{om} values for PAH may differ within an order between different soils [30]. Also, the sensitivity analysis showed that K_{om} has a large effect on the calculated biodegradation curves (Figure 6.2a). Independent experimental determination of K_{om} values will improve the predictive quality of the model.

Figure 6.1

Relative concentrations of three different PAH compounds in three different soil or sediments. The figures show modelled degradation (Δ), experimental degradation (×), and experimental desorption (○).
(a) phenanthrene in KGP
(b) phenanthrene in SWP
(c) phenanthrene in PHS
(d) chrysene in KGP
Error bars indicate 1 standard deviation for the experimental degradation results. Standard deviations for the experimental desorption results are too small to be shown.

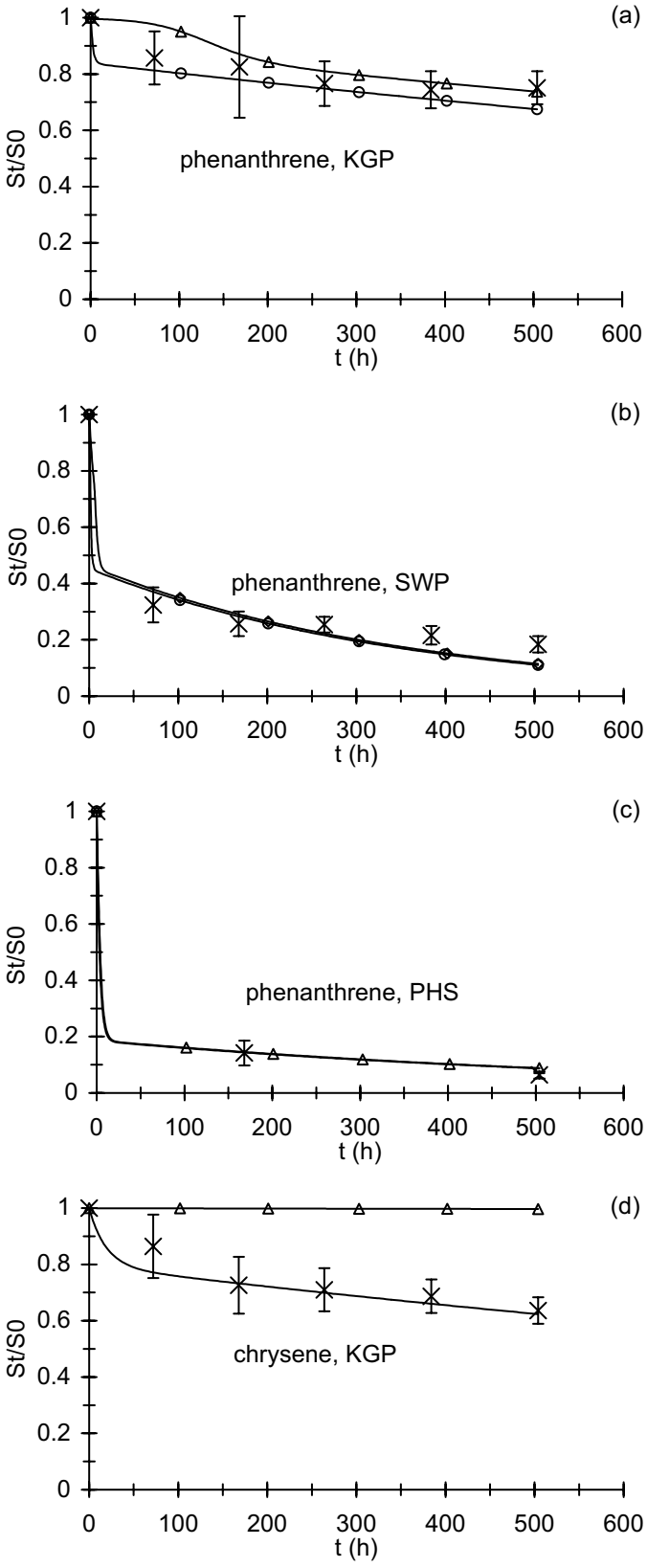
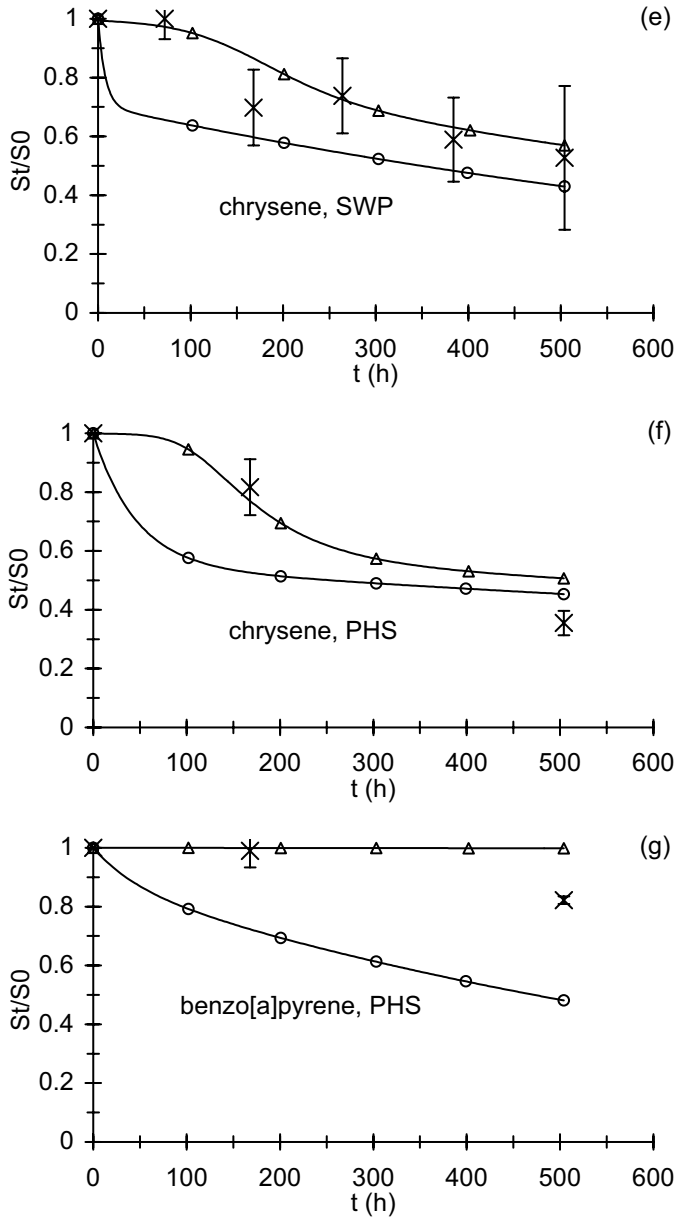


Figure 6.1 (continued)

(e) chrysene in SWP
(f) chrysene in PHS
(g) benzo[a]pyrene in PHS.



DEGRADATION RESULTS OF DIFFERENT PAH COMPOUNDS

Figures 6.1a-c showed that the calculated and experimental biodegradation rates of phenanthrene were high and very similar to the maximal desorption rates. This implies that desorption and not biodegradation was limiting removal of phenanthrene from soil. For chrysene (Figures 6.1d-f), the initial degradation was very slow, but, after about 50-100 hours, biodegradation rates increased and at the end biodegradation rates of chrysene were similar to the maximal desorption rates. This implies that at the start of the experiments biodegradation limited removal of chrysene, whereas at the end desorption limited removal.

Figure 6.2

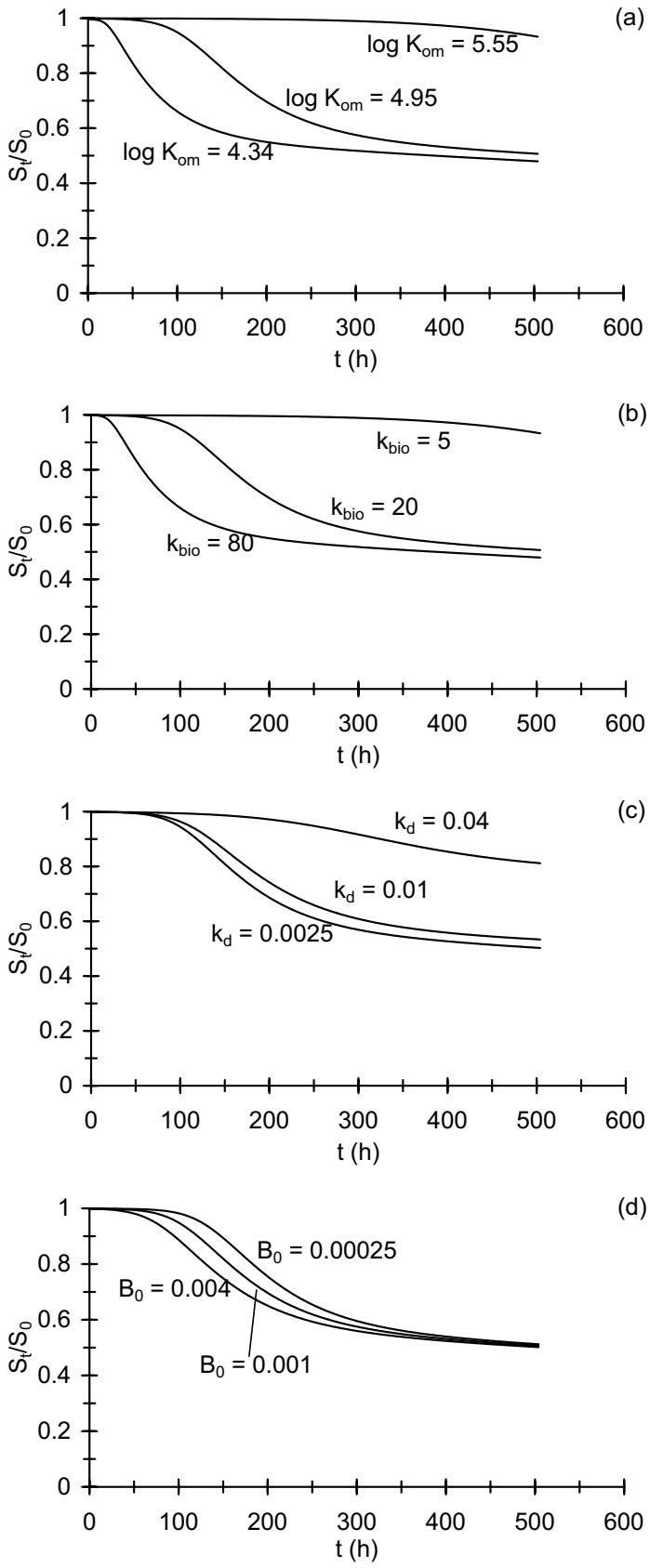
Sensitivity analysis for modelled degradation of chrysene in PHS.

(a) organic matter-water partition coefficient ($l \cdot kg^{-1}$)

(b) first order rate constant ($l \cdot mg^{-1} \cdot h^{-1}$)

(c) decay coefficient (h^{-1})

(d) initial biomass concentration (mg/l)



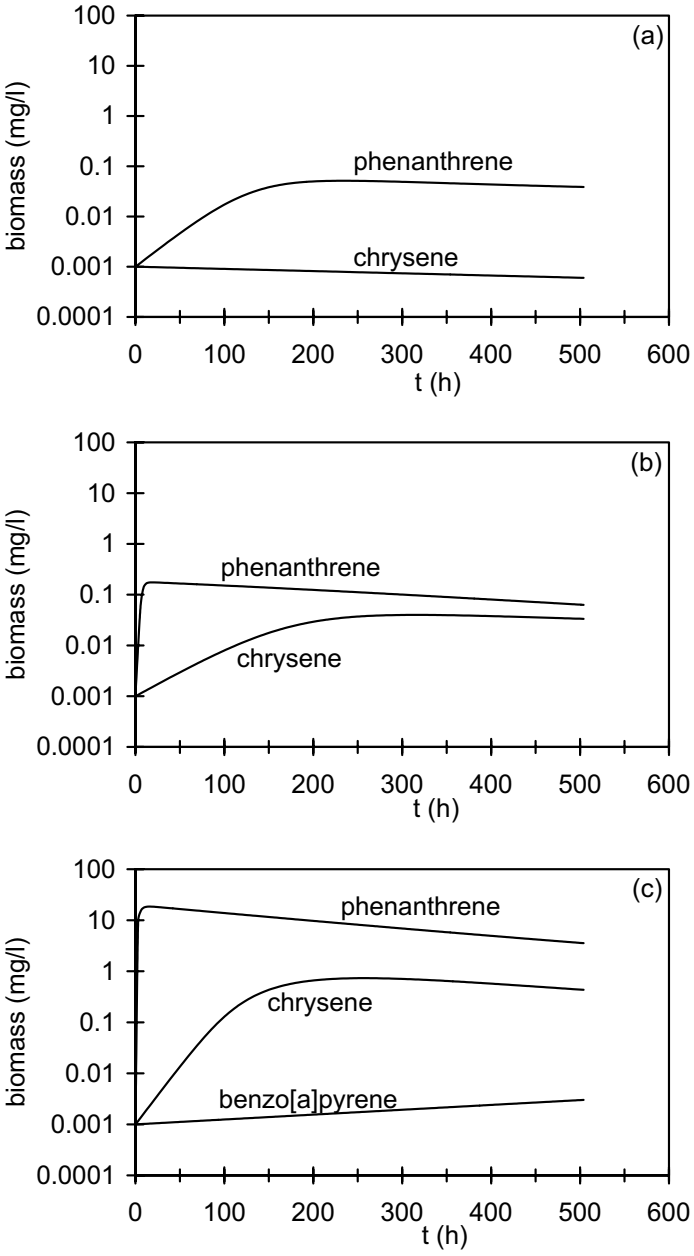
Calculated and experimental biodegradation rates of benzo[a]pyrene in PHS (Figure 6.1g) were always much lower than desorption rates. Removal of benzo[a]pyrene was therefore limited by biodegradation and not by desorption. The reason for this difference in behaviour between phenanthrene, chrysene, and benzo[a]pyrene is shown in Figure 6.3. These figures show the biomass concentrations during biodegradation, which were calculated with the model. Figure 6.3b and 6.3c show that the biomass concentration in SWP and PHS increased very fast for phenanthrene, which led to a biodegradation capacity that was much larger then the maximal desorption rate. Consequently, biodegradation rates followed the maximal desorption rates. For chrysene, biomass increased much slower and therefore, biodegradation was initially much slower compared to phenanthrene degradation. But eventually biodegradation of chrysene followed desorption, when the biomass concentration has reached its maximal value. The growth of the biomass that uses benzo[a]pyrene (Figure 6.3c) was very slow and biomass concentrations remained very low. As a result, biodegradation was very slow during the complete degradation time. The differences in the biomass growth rates were caused by the differences in equilibrium concentrations in the aqueous phase for the different PAH compounds. Consequently, the highest equilibrium concentrations (here phenanthrene) led to the highest growth rates of the biomass.

REMEDIATION OF HIGH MOLECULAR WEIGHT PAH

These results clearly show that high molecular weight PAH, like benzo[a]pyrene, can not serve as growth substrates for micro-organisms in soil, despite of the relatively high concentrations of these compounds that were observed for PHS in this study. This probably is the reason for the persistence of these compounds in soil systems. It can then be understood why so far no bacteria have been isolated which can grow on high molecular weight PAH as primary substrate [1,2].
However, Figure 6.1g shows that in the experimental biodegradation studies and also several other studies on PAH biodegradation in soil (e.g. [1,7]), limited degradation of benzo[a]pyrene and other high molecular weight PAH was found. This removal is probably the result of co-oxidation processes of these high molecular weight PAH by bacterial strains growing on smaller PAH compounds. Various studies showed that several bacterial strains that grow on 2, 3, or 4 ring PAH can cometabolise one or more 5-6 ring PAH compounds to some extent [1,20,31].

Figure 6.3

Modelled biomass concentrations during biodegradation of phenanthrene, chrysene and benzo[a]pyrene in
(a) KGP soil
(b) SWP soil
(c) PHS soil.



Cometabolism rates are generally much lower than degradation rates of primary substrates, which corresponds with the slow removal of benzo[a]pyrene in this study. It is unlikely that a too high value of K_{om}, used in the model, is the reason for the difference between experimental and modelled results of benzo[a]pyrene in PHS, like for phenanthrene and chrysene degradation in KGP. Because a lower K_{om} of benzo[a]pyrene implies lower K_{om} values for phenanthrene and chrysene in PHS too, and phenanthrene and chrysene degradation can be described well by the model.

CONCLUSIONS

A coupled sorption and biodegradation model with biphase sorption kinetics and Monod-kinetics for biodegradation can describe bioremediation results of PAH contaminated soil well. The model showed that persistence of high molecular weight PAH in soil systems can be caused by the very low aqueous phase concentrations of these compounds. These concentrations nearly meet the maintenance requirements of biomass degrading these substances and consequently lead to very slow growth of the biomass. However, co-oxidation of these high molecular weight PAH may lead to a larger removal during experiments than is predicted by the model. The modelling results were most sensitive to the soil-water partition coefficient of the contaminants and the decay rate of the biomass. Especially measurements of the partition coefficients for the different soils will further improve the predictive value of the model.

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7

Cost Aspects of Thermal Pretreatment to improve Bioremediation

Previous experiments have shown that short-term heating of soil contaminated with PAH compounds reduces residual concentrations after subsequent bioremediation. Different techniques for thermal pretreatment in several bioremediation processes for contaminated soils were discussed. Treatment costs of the different pretreatment techniques were calculated using a factorial cost estimate method. The combined costs of thermal pretreatment and subsequent bioremediation were compared to costs of alternative strategies for remediation. It was found that thermal pretreatment can be incorporated successfully in current bioremediation practice on both technical and economical grounds. Especially, the use of steam injection in biopile remediation and at in-situ remediation is a competitive alternative to current bioremediation techniques.

INTRODUCTION

For soils contaminated with organic compounds a wide spectrum of decontamination techniques can be used. Decontamination techniques can be divided in ex-situ techniques, in which soils are excavated before decontamination, and in-situ techniques, in which soil is not excavated and left in place during decontamination. Both excavated and non-excavated soils can be remediated using physical/chemical techniques and biological techniques. For ex-situ decontamination the most applied physical/chemical techniques are thermal decontamination (i.e. evaporation and incineration of the contaminants) and soil washing/classification. During soil washing/classification, contaminated soil is separated in a clean sandy fraction and a highly contaminated sludge fraction that has to be treated or disposed. The major advantage of physical/chemical remediation, compared to bioremediation, is the complete removal of contaminants. Bioremediation, which is generally cheaper and less energy consuming, can often not decrease contaminant concentrations below legal standards. Besides remediation, large volumes of excavated soils are disposed at controlled disposal sites.

In chapter 3 of this thesis it has been shown that short thermal pretreatment of PAH contaminated soil and classification sludge leads to an increase of the biodegradable fraction of the contaminants and consequently a decrease of the residual concentrations after bioremediation. Experiments described in chapter 4 showed that this increase of the biodegradable fraction was caused by an increase in the contaminant fractions that exhibited fast desorption behaviour. In that chapter, the effects of different pretreatment times, pretreatment temperatures, and water contents during thermal pretreatment were studied also. It was found that longer pretreatment times and higher pretreatment temperatures lead to larger fractions of the contaminants that exhibit fast desorption and, consequently, to lower residual concentrations that can be reached by bioremediation.

In case a combination of thermal pretreatment and bioremediation can reduce contaminant concentrations below legal standards and bioremediation itself can not, this combination might be an alternative for physical/chemical remediation. In this chapter the technological and economical possibilities for application of a thermal pretreatment in soil bioremediation are studied. Different techniques for thermal pretreatment in several soil bioremediation processes will be discussed and costs of the techniques will be calculated. The costs of combined thermal

pretreatment and subsequent bioremediation will be compared to costs of alternative strategies for remediation of soil and classification sludge.
An aspect that has to be considered when discussing the application of a thermal pretreatment, is the effect of a thermal pretreatment on the soil microflora. This is of importance, because the soil microorganisms perform the actual decontamination. Heating of soil to 100 °C will most certainly lead to sterilisation of the soil and removal of the soil's bioremediation potential. Heating soil to temperatures above 40 °C can lead to a decrease of biological activity and numbers of microorganisms [1,2]. It has even been found that the use of radio-frequency waves for soil heating decreased the microbial population for temperatures below 40 °C [3]. Consequently, microorganisms have to be re-introduced to thermally treated soil. This re-inoculation will be considered when discussing the thermal pretreatment techniques.

ECONOMICAL EVALUATION METHOD

EVALUATED REMEDIATION STRATEGIES

To determine the economical feasibility of a thermal pretreatment for different remediation strategies three aspects need to be considered. First, the costs for the application of a thermal pretreatment are calculated. The most important costs will be capital costs and energy costs, as heating of soil is a process consuming much energy. Second, the costs of the remediation technologies without the application of a thermal pretreatment are determined. Finally, the combined costs of pretreatment and bioremediation need to be lower or at least comparable to costs of competitive remediation strategies. For soils contaminated with hydrophobic organic compounds these alternatives are disposal of contaminated soil and thermal decontamination. Soil washing/classification can be a preliminary process to every remediation process that is discussed in this chapter. The classification sludge that is produced with this technology can be disposed or remediated using biological or physical/chemical techniques. Soil washing/classification is not included in the evaluation process.
The application of thermal pretreatment is evaluated on its technical and economical aspects for two different remediation strategies. These two strategies are currently applied without thermal pretreatment in full-scale processes for soils contaminated with hydrophobic organic compounds. The two remediation techniques are (a) decontamination in slurry reactors, (b) treatment using biopiles. In-situ bioremediation of contaminated soil is not included because of

the large variety in technologies that can be used for in-situ remediation. Further, the costs of the remediation itself do not entirely determine the choice for in-situ remediation. Very often additional costs like excavation of the soil and demolishing buildings, roads, etc. determine the choice between in-situ or ex-situ remediation. However, it can be assumed that costs of application of thermal pretreatment for in-situ remediation do not differ much from application in biopiles.

PROCESS CONDITIONS

The cost calculations were made for a thermal pretreatment at 100 °C during 1 to 24 hours. Experimental results have shown that lower temperatures are much less effective (Chapter 4 of this thesis). Pretreatment temperatures higher than 100 °C lead to pressures above 1 atmosphere and, consequently, require special and more expensive equipment for thermal pretreatment.

For the cost calculations of the slurry reactors and the biopiles, the following assumptions were made: 5000 tons dry solids per year are treated, the remediation process runs 7000 hours per year (80 % on-line), the thermal pretreatment step is added to an existing remediation process, which reduces the cost for labour, utilities, and buildings. The dry solids that are treated can be both excavated soil and classification sludge from a soil washing plant.

METHOD OF COST ESTIMATION OF THERMAL PRETREATMENT

To calculate the costs of a thermal pretreatment, the capital costs and the operating costs have to be estimated. The capital costs determine the investment for the addition of a thermal pretreatment step to a bioremediation process. The operating costs are the annual costs for operating the pretreatment process.

Capital costs include the pretreatment equipment, the installation of the equipment, and engineering costs. Capital costs were calculated using a so-called 'factorial cost estimate', in which equipment costs (EC) are multiplied with appropriate factors for direct costs (piping, instrumentation, electrotechnics, buildings) and indirect costs (engineering, contracting, contingency) [4]. Because a thermal pretreatment was regarded as an extension of an existing remediation process, some direct costs like storage, site development, and utilities could be omitted.

The equipment costs consist of the costs of all major process equipment. They were estimated using recent or historical data from cost engineering handbooks, papers, or reports. All prices and costs that are mentioned in this chapter are 1999 prices. When no 1999 prices could be found, prices were recalculated from original data using price indices [5].

The applied cost factors for calculation of the direct costs were: equipment erection (f=0.4), piping (f=0.7), instrumentation (f=0.2), electrical (f=0.1), process buildings (f=0.15). The sum of equipment costs and direct costs yields the physical plant costs (PPC).
The applied cost factors for the indirect costs were: design and engineering (f=0.3), contractor's fee (f=0.05), and a contingency allowance to cover unforeseen circumstances (f=0.1). The sum of the indirect costs and the physical plant costs yields the total fixed capital (FC).
Operating costs were divided in fixed costs, which do not vary with the amount of soil treated, and variable costs, that are dependent on the amount of soil treated. The operating costs were calculated on an annual basis.
The fixed costs include maintenance, tax and insurance, labour, and the capital charge. The capital charge was calculated on basis of the fixed capital using an operating life of 10 years and a 10 % interest rate. Then the annual capital charge is 16.5 % of the fixed capital. The maintenance costs and the tax plus insurance were estimated to be 5 % and 2 % of the fixed capital, respectively. Labour costs will be low or zero, because a thermal pretreatment is regarded as an extension of an existing remediation process.
The variable costs include costs for utilities (e.g. steam, oil, water, electricity) and costs for miscellaneous materials that are not covered by maintenance or utility costs. Utilities are mainly determined by the energy requirements for heating of the soil. Costs for miscellaneous materials were estimated to be 5 % of the maintenance costs.

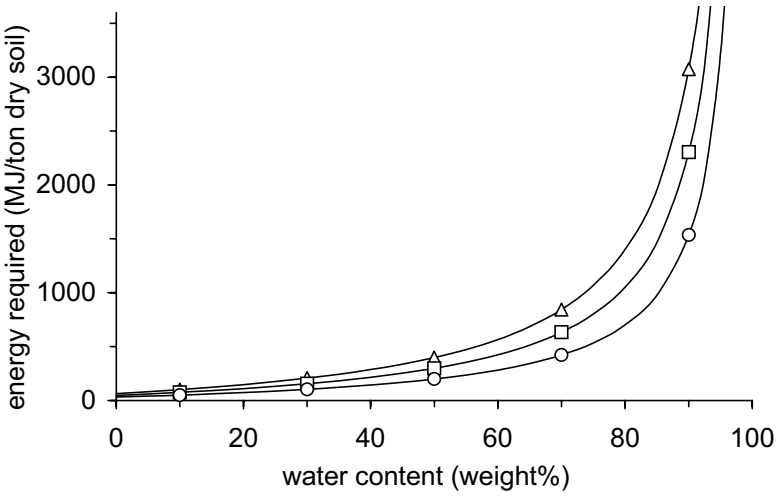
COST ESTIMATES OF THERMAL PRETREATMENTS

ENERGY REQUIREMENTS FOR THERMAL PRETREATMENT

The energy required for raising the temperature of soil depends mainly on the water content of the soil, as the specific heat of water (4.18 $kJ \cdot kg^{-1} \cdot K^{-1}$) is 5 times as high as the specific heat of dry soil material (approximately 0.8 $kJ \cdot kg^{-1} \cdot K^{-1}$) [6]. The required energies for heating soil from 20 °C to 60, 80, or 100 °C are shown in Figure 7.1 as function of the water content. This figure shows that the energy requirements increase almost exponentially with increasing water content. Typical water contents are: 10-30 weight% for terrestic soils, 30-60 weight% for dredged sediments, and 80-90 weight% for soil slurries. In general, water contents are higher for clay and peat soils than for sandy soils.

Figure 7.1

Energy requirements as function of the water content for heating soil from 20 °C to three different temperatures
○ 60 °C
□ 80 °C
Δ 100 °C



TREATMENT OF SOIL IN SLURRY REACTORS

In soil-slurry remediation, a soil-water mixture with a solids content of 10 to 20 weight percent is intensively mixed and aerated to prevent mass transfer limitations of the contaminants, nutrients, and oxygen and to obtain maximal biodegradation rates. Soil slurry remediation can very well be applied for fast reduction of relatively high contaminant concentrations. Currently, slurry remediation is not applied in the Netherlands, although several pilot scale studies have been conducted [7,8]. In the United Stated there are both pilot-scale and full-scale installations for soil slurry remediation [9,10].

In general, the re-inoculation problem as discussed previously will not occur when using slurry reactors. Currently, slurry reactors are operated either as continuous systems or as fed batch systems, in which the contaminated sludge is inoculated with a small volume of previously decontaminated sludge that contains high numbers of contaminant degrading microorganisms. In a fed batch system the pretreated slurry needs to be cooled before inoculation, because the slurry temperature is too high for biodegradation immediately after pretreatment. In a continuous system, the large reactor volume will buffer the high temperature of pretreated slurry.

The costs for adding a thermal pretreatment step to a slurry remediation process are very much determined by the energy costs for heating, because of the low solids content of the slurry. Solids contents of a slurry are generally 10-20 weight%. This means that 1400 to 3100 MJ per ton dry solids is required for heating from 20 to 100 °C (see Figure 7.1). This is equivalent to a heat flow of 278 (20 % soil slurry) to 615 kW (10 % soil slurry) when treating 5000 ton dry soil per year. Cost calculations are made for two different heating technologies,

the first one with direct heating using a furnace (Figure 7.2a), the second one using low-pressure steam and a heat exchanger (Figure 7.2b and 7.2c). In the cost calculations of both processes, labour costs are not incorporated because both thermal pretreatment and soil slurry remediation are technological, semi-automated processes. Therefore, it is expected that the use of a thermal pretreatment, as being an additional unit operation, can be incorporated in normal process operation for slurry remediation without extra labour required. Capital costs and operating costs for the application of a thermal pretreatment in soil slurry remediation are presented in Table 7.1.

To calculate the costs of a thermal pretreatment using a furnace, the heat per unit time was required: 278 kW (20 % slurry) and 615 kW (10 % slurry), respectively. For calculation of the energy costs, a heating efficiency of 80 % was assumed.

Figure 7.2

Flow-sheets for soil slurry remediation with a thermal pretreatment by
a) furnace heating
b) heat exchanger
c) heat exchanger, with energy recovery using a second heat exchanger

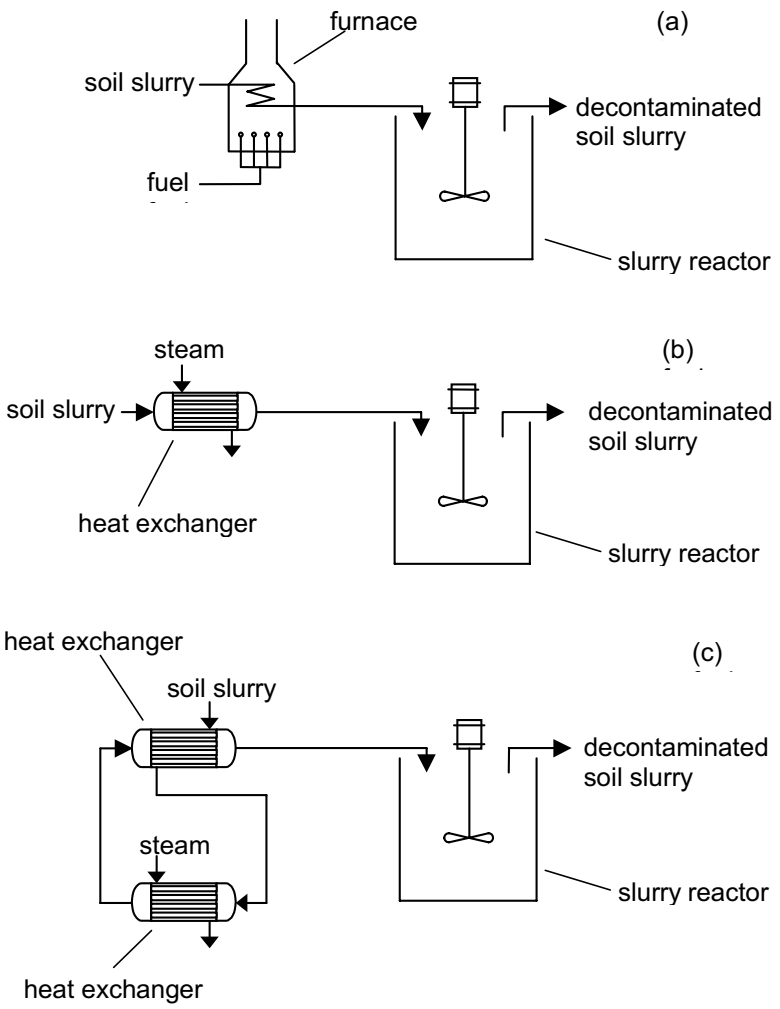


Table 7.1 Cost calculations for thermal pretreatment of soil slurries

	costs (k€)			
	furnace		heat exchanger	
	10 % slurry	20 % slurry	10 % slurry	20 % slurry
Capital				
equipment costs (EC)	40[a]	22[a]	18[b]	15[b]
physical plant costs (PPC: 2.55xEC)	102	56	45	38
fixed capital (FC: 1.45xPPC)	148	81	66	55
Fixed costs				
capital charge (0.165 x FC)	24.4	13.4	10.9	9.2
maintenance (0.05 x FC)	7.4	4.1	3.3	2.8
tax/insurance (0.02 x FC)	3.0	1.6	1.3	1.1
total fixed costs	34.8	19.1	15.5	13.1
Variable costs				
miscellaneous materials (0.05 x maintenance)	0.37	0.20	0.16	0.14
oil (H = 41.3 GJ/ton, price = 170 €/ton[b])	78.4	35.6		
steam (H_{cond} = 2.26 GJ/ton, price = 15 €/ton[b])			99	45
total variable costs	78.8	35.8	99	45
total operating costs	113.6	54.9	114.5	58.1
costs / ton d.s.	23 €/ton	10 €/ton	23 €/ton	12 €/ton

[a]from reference [4]
[b]from reference [5]

For calculating the costs of thermal pretreatment using a heat exchanger, first the size of the heat exchanger, this means the required heat transfer area, had to be

determined. The calculation of the heat transfer area of a heat exchanger is given in Appendix I of this chapter. Using a steam temperature of 120 °C, it can be calculated that the required heat transfer areas are 16 m^2 for a 10 % soil slurry and 7.5 m^2 for a 20 % soil slurry.
Table 7.1 shows that the costs for a thermal pretreatment are nearly identical for using a furnace or a heat exchanger, although fixed costs are more important when using a furnace. The costs for the heat exchanger are mainly determined by the energy requirements (i.e. steam), which determine almost 80-90 % of the total costs. Because the capital costs are relatively low compared to the energy costs, total costs can be reduced with 15 to 30 % by using an additional heat exchanger that regains energy from pretreated soil slurry (Figure 7.2c). The energy savings will most likely be larger than the costs of the additional heat exchanger. A disadvantage of this set-up might be a reduction of the pretreatment time of the soil, because the heated soil slurry is immediately cooled down in the additional heat exchanger. A shorter pretreatment time might reduce the pretreatment effect.

TREATMENT OF SOIL USING BIOPILES

Bio-piles and landfarms are more extensive treatment technologies for excavated soils compared to slurry treatment. In both biopiles and landfarms, excavated soil is spread in layers of 0.5 to 1.5 m. In biopiles, the soil is regularly tilled during the remediation process and nutrients and water contents are monitored and regulated using watering and drainage systems. Landfarming is a more extensive process, in which only pollutant concentrations are monitored.
The techniques for heating soil as a pretreatment step in biopiles, landfarms or at in-situ remediation is different than heating soil that is bioremediated in slurry reactors. As the soil is in solid state, no direct heating using furnaces or heat exchangers can be applied.
For in-situ remediation, which will not be discussed in detail further, heating of soil has already been applied to stimulate remediation by means of soil vapour extraction (SVE) of volatile and semi-volatile contaminants. Under the EPA Superfund program several full-scale and pilot-scale tests have been carried out to improve SVE using three different technologies: steam injection, radio-frequency heating, and electrical resistance heating [11-13]. In all tests, soil was heated to 100 °C or more. It was found that steam injection could only be used in well-permeable soils, thus sandy soils. On the other hand, radio-frequency heating and electrical resistance heating were most effective in heating clay soils than sandy soils, because radio-frequency energy is better absorbed and electric current is better conducted due to the higher water contents in clay and peat soils.

Radio-frequency heating has also been tested to stimulate bioremediation in arctic conditions by raising the soil temperature to ambient values [3,14,15].
Solid contents of soil in solid state, as in biopiles, are much higher than those of soil slurries. Typical contents are between 70 to 90 weight percent. This means that energy requirements for heating soil from 20 to 100 °C range from 100 to 210 MJ per ton dry solids (see also Figure 7.1). In contrast to slurry remediation, heat from pretreated soil can not be recovered easily.
Cost estimations are only made for thermal pretreatment in biopiles. The calculations are performed for thermal pretreatment by steam injection and by radio-frequency heating. In contrast to the slurry remediation, labour costs are incorporated for remediation with biopiles, as the addition of a thermal pretreatment step to a more extensive remediation process, like biopiles, is likely to require additional labour.

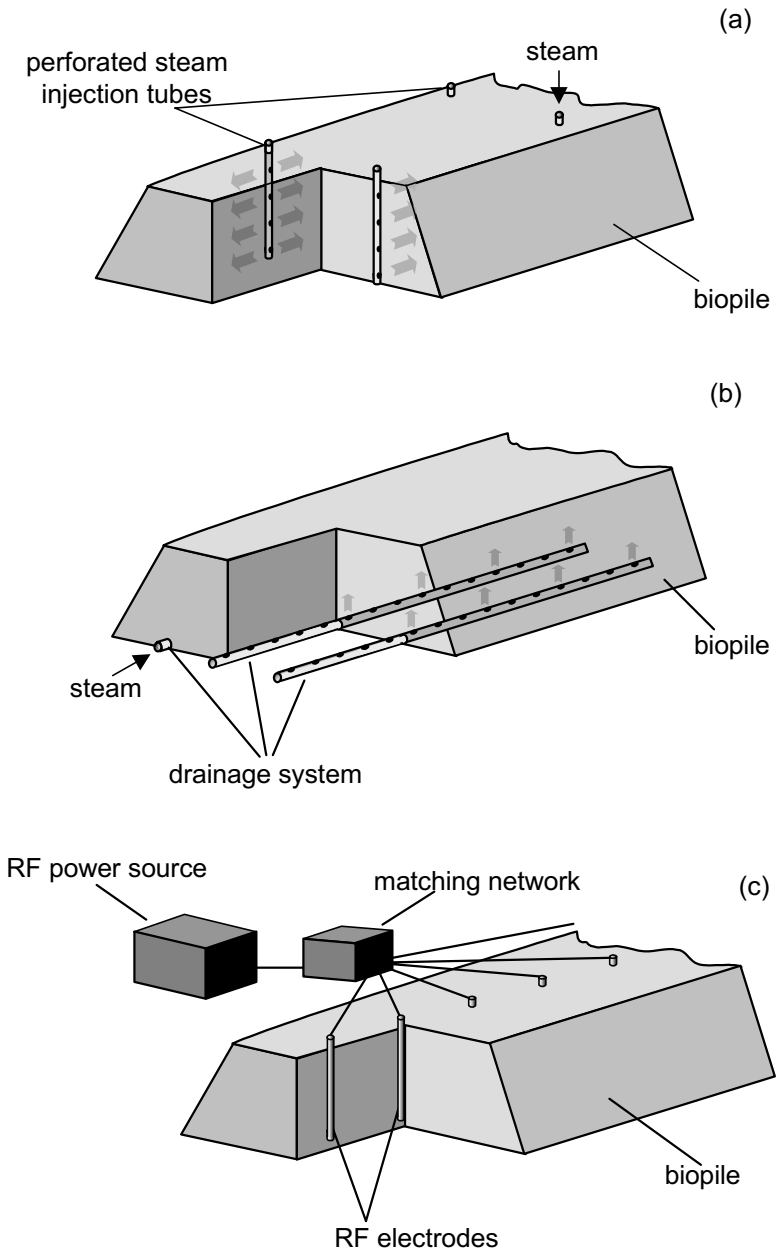
For steam injection, capital costs are negligible, because steam itself is considered as a utility and steam injection can be performed using simple perforated stainless steel tubes (Figure 7.3a). In case the biopile is equipped with a drainage system, the tubing of the drainage system might be used for steam injection (Figure 7.3b). Steam is allowed to be considered as a utility when it can be obtained from a nearby power plant or chemical plant. In case, steam can not be obtained from nearby industrial plants and it has to be produced on-site, energy costs are estimated to be no more than twice as high. Precautions have to be taken in case the contamination also contains volatile or semi-volatile compounds that might evaporate from the soil during pretreatment. Then, air treatment systems (e.g. activated carbon filters or biofilters) have to be installed. These are not incorporated in the cost calculations
In case of radio-frequency heating, capital costs are a major part of the total costs. Capital costs are calculated for 70 kW (90 % solids) and 150 kW (70 % solids) systems. This means that 20 ton dry solids can be pretreated on an 8 hour working day, and thus 5000 ton per year. Capital costs include a RF power source (half of total capital costs), a control system, a matching network to supply the RF power to the electrodes, electrodes, and a RF shield to protect the environment from the magnetic field (Figure 7.3c).
Table 7.2 shows the cost calculations for a thermal pretreatment in biopiles using steam injection or radio-frequency heating. This table shows that costs for radio-frequency heating are much higher than using steam injection. This difference is mainly caused by the high capital costs for radio-frequency heating.
An alternative technique for heating soils with a low permeability can be electric resistance heating. Cost data for this technique could not be found, but much

lower costs were reported for the application of electric resistance heating to enhance soil vapour extraction than using radio-frequency heating [16]. This means that electric resistance heating might be a more feasible alternative for biopile heating.

Re-inoculation of thermally treated biopiles can be performed by mixing the thermally treated soil with a small amount non-treated soil that contains contaminant degrading micro-organisms.

Figure 7.3

Biopile heating by
a) steam injection using injection tubes
b) steam injection using a drainage system
c) radio-frequency heating



In case of in situ remediation, recolonisation of a thermally treated area may take place via groundwater flow from adjacent areas that have not been pretreated, without any explicit addition of micro-organisms.

Table 7.2 Cost calculations for thermal pretreatment of soil in biopiles

	costs (k€)			
	steam injection		radio frequency heating	
	70% solids	90% solids	70% solids	90% solids
Capital				
equipment costs (EC)	0	0	no data	no data
physical plant costs (PPC: 2.55xEC)	-	-	838[a]	538[a]
fixed capital (FC: 1.45xPPC)	-	-	1214	780
Fixed costs				
capital charge (0.165 x FC)	-	-	200	129
maintenance (0.05 x FC)	-	-	61	39
tax/insurance (0.02 x FC)	-	-	24.3	15.6
labour (400 hrs, 30 €/hr[b])	12.0	12.0	12.0	12.0
total fixed costs	12	12	297	196
Variable costs				
miscellaneous materials (0.05 x maintenance)	-	-	3.04	1.95
steam (E_{cond} = 2.26 GJ/ton, price = 15 €/ton[b])	6.7	3.3	-	-
electrical power (16.5 €/GJ[b])	-	-	17.3	8.3
total variable costs	6.7	3.3	20.3	10.25
total operating costs	18.7	15.3	317	206
costs / ton d.s.	3.7 €/ton	3.1 €/ton	63.4 €/ton	41.2 €/ton

[a]from reference [17]; costs were reported as physical plant costs instead of equipment costs

[b]from reference [5]

BIOREMEDIATION AND ALTERNATIVE TECHNOLOGIES

BIOREMEDIATION TECHNOLOGIES

The costs of the thermal pretreatment have to be added to the costs of the subsequent bioremediation technology to obtain an estimate of the total costs of soil remediation. Typical cost for bioremediation using slurry-reactors are between €60,- and €90,-/ton dry solids [18-20]. Costs for remediation of soil using biopiles are between €18,- and €45,-/ton dry solids [20,21].

ALTERNATIVE REMEDIATION TECHNOLOGIES

In 1998, 1900 ktons of excavated soil were decontaminated in the Netherlands, of which 700 ktons were decontaminated using thermal decontamination, 820 ktons by soil washing/classification, and only 380 ktons were biologically decontaminated [22]. Soil washing/classification is the most applied decontamination technology, however it produces a highly contaminated sludge fraction that still has to be treated by either thermal decontamination or bioremediation or has to be disposed. Therefore, soil washing/classification is not considered as an alternative technique for bioremediation, but only as a possible preliminary step.

The second largest technique for soil decontamination is thermal decontamination. During thermal decontamination, soil is heated to temperatures between 400 and 800 °C, depending on the type of contaminant. At these temperatures, organic contaminants evaporate from the soil or decompose; the evaporated contaminants are incinerated in a subsequent step. Costs for thermal pretreatment of contaminated soil are relatively high because of the large energy requirements for heating of the soil. Costs were estimated to range from €50,- to €80,- per ton [23]. A different report, comparing different thermal decontamination techniques, showed higher costs (€77,- to €115,- per ton), but the revenues of the soil product after decontamination were not incorporated here [24].

Another alternative for bioremediation of contaminated soil is disposal at controlled disposal sites. Prices for disposal differ widely for different disposal sites and different provinces in the Netherlands [25]. Prices for disposal of slightly contaminated soil, with contaminant concentration below the Dutch intervention value (I-value), differ from €14,- to €36,- per ton, with an average of €23,- per ton. For more heavily polluted soils, with concentrations above the I-value, prices differ from €36,- to €82,- per ton, with an average of €50,- per ton. In general, prices for disposal are the highest for the provinces in the east, south,

and north of the Netherlands, whereas they are the lowest for the provinces in the west (Noord-Holland, Zuid-Holland, and Utrecht).

DISCUSSION AND CONCLUSION

Table 7.3 shows a summary of costs for thermal pretreatment, biological remediation, and alternative remediation technologies.
This table shows that costs of bioremediation techniques without pretreatment are the lowest when using biopiles. The application of soil slurry reactors for decontamination is the most expensive technique. However, as biological decontamination can often not reduce contaminant concentrations below legal standards, soil has to be disposed, which is the second cheapest technique, or thermally decontaminated.

Table 7.3 Summary of cost estimates for thermal pretreatment, biological remediation and alternative strategies

remediation and pretreatment technology	costs (€/ton d.s.)[a] pretreatment	bioremediation	total
Slurry reactors			
furnace heating	10-23	60-90	70-113
heat exchanger	12-23	60-90	72-113
Biopiles			
steam injection	3.1-3.7	18-45	21-49
radio-frequency heating	41-63	18-45	59-109
Alternative strategies			
thermal decontamination	-	-	50-80
disposal	-	-	36-82[b]

[a]costs are 1999 prices, except for disposal (1998 prices)
[b]prices for soil with contaminant concentrations above Dutch intervention-value

To increase the amount of soil that can be bioremediated to contaminant concentrations below legal standards, a thermal pretreatment step can be added to a bioremediation process. The use of steam injection in biopiles is the cheapest techniques and prices are lower or comparable to alternative remediation methods. The use of radio-frequency heating in biopile remediation yields an expensive technique, because of very high capital costs. Also, the addition of a thermal pretreatment to soil slurry remediation yields an expensive technique that is not competitive to disposal and thermal decontamination. The pretreatment costs in slurry remediation are relatively high, because of the high energy costs for heating a soil slurry; although regaining part of the energy might reduce these costs. Further, the use of slurry reactors is very expensive itself.
For in-situ remediation, it can be expected that the costs for a thermal pretreatment do not differ much from the costs of a thermal pretreatment in biopiles. Consequently, the application of steam injection during in-situ remediation might be feasible too.
It should be noted that the application of thermal pretreatment can reduce the residual concentrations after subsequent bioremediation, but that these residual concentrations are not necessarily below legal standards. This has to be examined for each contaminated soil individually.

Summarising, a combination of thermal pretreatment and subsequent bioremediation of contaminated soil leads to a remediation system that is technically and economically feasible and can be incorporated in current bioremediation practice. Especially, the use of steam injection in biopile remediation can yield a competitive alternative to current remediation techniques.

APPENDIX I. CALCULATION OF THE HEAT TRANSFER AREA OF A HEAT EXCHANGER

The amount of heat that can be transferred per unit time by a heat exchanger is given by [4]:

$$\Phi_w = UA\Delta T_m \tag{1}$$

where Φ_w is the heat transferred per unit time (W); U is the overall heat transfer coefficient (W/m^{2}K); A is the heat transfer area (m^2); ΔT_m is the logarithmic mean temperature difference (K). The value of U is determined by the partial heat transfer coefficients of the flows outside and inside the tubes of the heat exchanger (here: steam and soil slurry), corrected for fouling of the tubes, and by the thermal conductivity of the tube wall.

$$\frac{1}{U} = \frac{1}{\alpha_o} + \frac{1}{\alpha_{fo}} + \frac{d_o \ln(d_o / d_i)}{2\lambda_w} + \frac{d_0}{d_i} \cdot \left(\frac{1}{\alpha_i} + \frac{1}{\alpha_{fi}} \right) \tag{2}$$

where α_o, α_i are the heat transfer coefficients inside and outside the tubes, respectively (W/m^{2}K); α_{fo}, α_{fi} are the fouling coefficients inside and outside the tubes (W/m^{2}K); d_o, d_i are the outside and inside diameters of the tubes (m); λ_w is the thermal conductivity of the tube wall material (W/mK).
In general, the flow that shows the greatest risk of fouling of the heat exchanger will be sent through the tubes. In this case, the slurry, containing solid particles, gives the greatest risk of fouling of the heat exchanger. The heat transfer coefficient for flow of a fluid through tubes can be calculated using:

$$Nu = 0.023 Re^{0.8} Pr^{0.33} \tag{3}$$

where

$$Nu = \frac{\alpha_i d_i}{\lambda} \tag{4}$$

$$Re = \frac{\rho v d_i}{\eta} \tag{5}$$

$$Pr = \frac{C_p \eta}{\lambda} \tag{6}$$

and λ is the thermal conductivity of the fluid (W/mK); ρ is the density of the fluid (kg/m^3); v is the fluid velocity (m/s); η is the fluid viscosity (Pa·s); C_p is the fluid heat capacity (J/kgK).

The maximal fluid velocity for flow through tubes is 2.5 m/s for aqueous fluids. This maximal velocity should be used to reduce fouling. Values for α_i of 8000 W/m^{2}K can be calculated using the following values for the constants in equations 3-6: d_i is 16 mm (standard ¾ in. tube), λ is 0.6 W/mK; ρ is $1.07 \cdot 10^3$ kg/m^3, η is $1 \cdot 10^{-3}$ Pa·s; C_p is 3.67 kJ/kgK. Typical values for α_o (steam condensation), α_{fi}, and α_{fo} are 8000, 3000, and 5000 W/m^{2}K, respectively. A value of 960 W/m^{2}K for U can be calculated using λ_w is 16 W/mK (stainless steel) and d_o is 20 mm (standard ¾ in. tube).

The logarithmic mean temperature difference (ΔT_m) can be calculated using the following equation:

$$\Delta T_m = \frac{T_{slurry,out} - T_{slurry,in}}{\ln\left(\frac{T_{steam} - T_{slurry,in}}{T_{steam} - T_{slurry,out}}\right)} \qquad (7)$$

where $T_{slurry,\ out}$ is the temperature of the soil slurry leaving the heat exchanger (K); $T_{slurry,in}$ is the temperature of the soil slurry entering the heat exchanger (K); T_{steam} is the temperature of the steam that is used to heat the slurry (K).

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8

General Discussion

INTRODUCTION

The objective of the study presented in this thesis was to investigate methods that might improve bioremediation of PAH contaminated soils, i.e. to decrease residual concentrations after bioremediation. From a literature study it was concluded that al methods that were directed to an increase of the pore diffusion rate, like breaking up of soil aggregates or the use of surfactants, could increase initial degradation rates but could not reduce residual concentrations. Consequently, it was concluded that only methods that could decrease sorption of contaminants to soil organic matter might decrease residual concentrations. Soaking with an organic solvent and short term heating were selected as the most promising techniques to decrease sorption of contaminants and, thus, to improve subsequent bioremediation. This was confirmed in a laboratory study, in which was shown that both techniques increased PAH bioavailability and consequently led to lower residual concentrations after biodegradation. Thermal pretreatment showed the best results and was selected for further study, also because the implementation of soaking with a high volatile solvent in current bioremediation practice would require extensive and expensive safety measurements.

The effects of thermal pretreatment on PAH desorption kinetics were studied and it was found that part of the soil sorbed PAH contaminants exhibit faster desorption after a short thermal pretreatment of the contaminated soil. Prolonged pretreatment times and higher pretreatment temperatures could even further enlarge the fast desorbing contaminant fractions. It was then concluded that this improved desorption leads to a greater bioavailability of the PAH contaminants and consequently to improved bioremediation results.

Once, it had been shown that a thermal pretreatment is an effective technique to improve bioremediation, it was necessary to determine whether it is technically and economically feasible to incorporate a thermal pretreatment in current bioremediation practice. An economical en technical evaluation showed that a combination of thermal pretreatment and subsequent bioremediation of contaminated soil can lead to a remediation system that is a competitive alternative to current remediation techniques, such as incineration or disposal.

It can be concluded that a potentially successful technique has been developed for improved bioremediation of PAH contaminated soils. However, there are a few points that require further discussion. First, so far the effects of thermal pretreatments have only been tested in slurry systems on a few soil types. Further, PAH contaminants were the only compounds studied. Hence, it will now be discussed in which bioremediation systems a thermal pretreatment can be

incorporated. It will also be discussed for what soil types and contaminant species a thermal pretreatment can increase bioavailability. Some special attention will be given to high molecular weight PAH, because these compounds showed to be very persistent to biodegradation, even after thermal pretreatment.
Further, policy and legislation towards soil contamination and decontamination have changed dramatically during last years and are still changing. Hence the roles of pretreatments in current and future remediation practices will be discussed.

APPLICABILITY OF THERMAL PRETREATMENT

APPLICABILITY FOR DIFFERENT BIOREMEDIATION TECHNOLOGIES

Experiments with thermal pretreatments, as described in Chapter 3 and 4, have only been performed in slurry systems. However, thermal pretreatment might also increase bioavailability using different bioremediation technologies, like biopiles.
Koning et al. [1] showed that residual concentrations of bioremediation in slurry systems are comparable to bioremediation in biopiles. Further, in Chapter 5 it has been shown that results of slurry phase bioremediation are determined by the fact that a fraction of soil sorbed contaminants exhibits slow desorption. These slowly desorbing fractions are comparable to the residual fraction after bioremediation. This corresponds with studies from Cornelissen et al. [2] who found that slurry phase desorption studies can be used to predict bioremediation results from biopiles. Together, all of these three studies showed that residual concentrations are independent of the applied system and that these results are determined by the same mechanism, namely slow desorption. Since a thermal pretreatment decreases the fraction of the contaminants that exhibits slow desorption, as has been shown in Chapter 4, it can be stated that this pretreatment will lead to similar increases in bioavailability for different bioremediation systems.

APPLICABILITY FOR DIFFERENT SOIL TYPES AND CONTAMINANT SPECIATIONS

Most experiments with thermal pretreatments, as described in Chapters 3 and 4, have been performed with classification sludge having a high clay content and a high organic matter content. As a result of the high organic matter content of this soil, most PAH contaminants will most likely be sorbed to the soil organic matter. The increase of the bioavailability in this soil after a thermal pretreatment

has been explained as a structural rearrangement of the soil organic matter (Chapter 4).
Some experiments described in Chapter 3 were performed with a different type of soil, which had a low clay content and low organic matter content. In this soil also tar particles were present. Most contaminants will then be sorbed inside or at the surface of the tar particles. For this soil a thermal pretreatment did not increase the bioavailability of the PAH contaminants.
These results from experiments with both soils clearly demonstrate that soil composition and, related to that, contaminant speciation are important factors concerning the effects of a thermal pretreatment. The results suggest that bioavailability of contaminants sorbed to natural soil organic matter can be increased by thermal pretreatment, whereas the bioavailability of contaminants sorbed in or at tar constituents can not. Consequently, thermal pretreatment might be best applied for high organic matter soils with no tar constituents. However, in this study a too limited number of soil types have been tested to fully validate this hypothesis.

APPLICABILITY FOR HIGH MOLECULAR WEIGHT PAH

Degradation of high molecular weight (HMW) PAH requires special attention because they are more persistent for degradation than low molecular weight (LMW) PAH.
An analysis of results from both desorption experiments and biodegradation experiments showed that desorption kinetics can be used to describe biodegradation kinetics of PAH compounds with two to four aromatic rings (Chapter 5). This confirms the general conception that bioremediation rates are determined by desorption rates. However, for larger PAH compounds (five and six aromatic rings) desorption was faster and to a greater extent than biodegradation, which means that slow biodegradation is responsible for the low biodegradation rates of these compounds.
Because desorption rates of HMW PAH exceed biodegradation rates by far, these compounds can accumulate in the water phase of the soil and, consequently, form a risk for toxic effects in the environment. Moreover, HMW PAH are the PAH compounds that include most of the carcinogenic PAH compounds. This means that effective detoxification of soils in which HMW PAH are abundant is difficult.

REMOVAL OF HIGH MOLECULAR WEIGHT PAH

In Chapter 5 it has been hypothesised that the persistence of HMW PAH in soil is caused by the low aqueous phase concentrations of these compounds. These

aqueous phase concentrations are too low to meet the maintenance requirements of the microorganisms degrading these substances and, as a result, growth of these microorganisms is very slow. This hypothesis was further supported by model calculations in Chapter 6, which indeed showed that no biomass could grow on HMW PAH only. Modelled biodegradation rates were actually lower than experimental rates, which was probably caused by some cometabolic degradation of these PAH in the biodegradation experiments. These findings are supported by literature data, which have reported only cometabolic degradation and no degradation of these PAH as a single carbon source so far [3].

To improve cometabolic degradation of HMW PAH, suitable compounds have to be added to the soil, which can serve as growth substrate for bacteria that can degrade HMW PAH as a secondary substrate. Several studies have been performed using compost as a growth substrate. Results, however, were ambiguous. Some authors have reported enhanced PAH degradation [4,5], whilst others have found no effect of compost addition [6].

Another approach for the removal of persistent organic compounds is the use of preliminary oxidation towards compounds that are more soluble, which can then be degraded by microorganisms. For oxidation of HMW PAH chemical techniques (advanced oxidation processes) [7] and biological means, for example with white-rot fungi, [8] have been studied. Both techniques have proved to be very effective in reducing PAH concentrations in spiked soils, although they could not increase bioavailability in aged soils (see also Chapter 2).

APPLICABILITY OF THERMAL TREATMENT FOR OTHER CONTAMINANTS

This study focussed on bioremediation of PAH contaminants. However, it has often been shown that residual concentrations and slow desorption are characteristic of all hydrophobic contaminants (e.g. [9]). The mechanism that determines this slow desorption is most likely very strong sorption of the contaminants to the soil organic matter (see also Chapter 1 and 4), although the exact causes of this strong sorption are still under discussion (e.g. [10,11]). Assuming that the mechanism of slow desorption is the same for different hydrophobic contaminants, it can be expected that a thermal pretreatment leads to an increase of the bioavailability of other contaminants than PAH as well.

PERSPECTIVES OF THERMAL PRETREATMENTS

PRETREATMENTS WITHIN CURRENT REMEDIATION PRACTICE

Currently, cleanup guidelines for soils contaminated with organic compounds are based on the total concentrations of these contaminants. This regards both the target and intervention values of the Soil Protection Act [12] and the composition values of the Construction Materials Decision [13]. This means that a thermal pretreatment can have an important function in remediation of soils for which guidelines can not be achieved using bioremediation under normal conditions. A thermal pretreatment might reduce residual concentrations after bioremediation to values below the cleanup guidelines. For sediments this can also mean that they end up in a different class of contamination, by which final processing or disposal is less expensive.

In addition, in Chapter 7 it has been shown that costs of thermal pretreatment can be relatively low compared to total remediation costs, depending on type of pretreatment technique and bioremediation technique. Also, combined costs of pretreatment and bioremediation were competitive to alternative remediation strategies as thermal decontamination or disposal.

PRETREATMENTS WITHIN FUTURE REMEDIATION PRACTICE

It can be prospected that future guidelines for organic contaminants will not merely be based on total concentrations but also on leaching levels. Currently, leaching levels are already in force within the Construction Materials Decision for guidelines of heavy metals. The use of leaching levels for organic contaminants means that removal of only the fast desorbing fraction of the contaminants in soil will mostly be sufficient to meet the contamination guidelines, because leaching from the slow desorbing fraction will be very small. Moreover, the use of leaching levels for organic contaminants is reasonable, as it has been shown that removal of the bioavailable fractions of soil sorbed contaminants largely reduces toxicity and plant uptake [14,15].

The use of a thermal pretreatment in bioremediation will generally not lead to less leaching but only to lower residual concentrations. In case of persistent contaminants, like HMW PAH, thermal pretreatment might even lead to increased leaching levels, due to an increase of the fast desorbing fraction. Altogether, the implementation of leaching based guidelines for PAH contaminated soils might decrease the demands for pretreatment technologies that increase bioavailability of soil sorbed contaminants.

RISKS OF AGED CONTAMINATIONS

One of the most important conclusions of this study is that once hydrophobic contaminants have sorbed strongly to soil, they can be released only very difficultly. Few pretreatments can increase desorption of these strongly sorbed contaminants. Moreover, the conditions that can lead to this increased desorption, like high temperatures or soaking with a solvent (Chapter 3), will not occur under natural environmental circumstances. It can then be concluded that risks of aged contaminations with PAH or other hydrophobic compounds are relatively small.

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Nomenclature

B	biomass concentration	$mg \cdot l^{-1}$
c	constrictivity	-
CMC	critical micelle concentration	$mg \cdot l^{-1}$
C_{surf}	aqueous surfactant concentration	$mg \cdot l^{-1}$
C_w	aqueous contaminant concentration	$mg \cdot l^{-1}$
D_b	diffusion coefficient in aqueous phase	$m^2 \cdot s^{-1}$
D_{eff}	effective diffusion coefficient	$m^2 \cdot s^{-1}$
F_{fast}	contaminant fraction exhibiting fast desorption	-
F_{slow}	contaminant fraction exhibiting slow desorption	-
f_{om}	organic matter content	-
k_{bio}	first-order rate constant for biomass growth	$l \cdot mg^{-1} \cdot h^{-1}$
k_d	biomass decay rate	h^{-1}
k_{fast}	first-order rate constant for fast desorption	h^{-1}
k_{slow}	first-order rate constant for slow desorption	h^{-1}
K_{mic}	micelles-water partition coefficient	$l \cdot kg^{-1}$
K_{om}	organic matter-water partition coefficient	$l \cdot kg^{-1}$
$K_{om,app}$	apparent organic matter-water partition coefficient in the presence of surfactants	$l \cdot kg^{-1}$
K_{ow}	octanol-water partition coefficient	-
K_p	soil-water partition coefficient	$l \cdot kg^{-1}$
K_s	affinity constant	$mg \cdot l^{-1}$
$K_{s \leftrightarrow f}$	equilibrium constant between fast and slowly desorbing compartment	-
m	maintenance coefficient	h^{-1}

r	distance from centre of soil aggregate	m
R	radius of soil aggregate	m
$r_{s,w}$	soil-water ratio	$kg \cdot l^{-1}$
S	soil sorbed concentration	$mg \cdot kg^{-1}$
S_{fast}	concentration of fast desorbing compounds	$mg \cdot kg^{-1}$
S_{slow}	concentration of slowly desorbing compounds	$mg \cdot kg^{-1}$
T	temperature	K
Y	microbial yield	-
ΔH_s	dissolution enthalpy	$J \cdot mole^{-1}$
ε	porosity	-
μ_{max}	maximal growth rate	h^{-1}
ρ	density	$kg \cdot m^{-3}$
τ	tortuosity	-

Summary

After the discovery of chemical waste in the soil under the village of Lekkerkerk in 1978, numerous sites in the Netherlands have been found to be contaminated. Approximately one percent of the contaminated sites contains polycyclic aromatic hydrocarbons (PAH). PAH contaminations of soil are of great concern since PAH can have toxic, mutagenic and carcinogenic effects. PAH are a group of hydrocarbons consisting of two or more fused benzene rings. They are non-volatile, they have very low solubilities in water and they tend to sorb very strongly to the solid soil matrix. As a result, remediation of PAH contaminated soil has proofed to be very costly and energy intensive.
Remediation of soil can be performed using physical/chemical techniques or biological techniques. Bioremediation has some major advantages above physical and chemical techniques, because with bioremediation the soil structure is preserved, no waste products are formed and energy requirements are generally low. However bioremediation of soils contaminated with PAH has often been unsuccessful due to slow degradation rates and high residual contaminant concentrations that do not meet legal standards. This slow and incomplete degradation is mainly caused by a slow desorption of PAH from the soil and not by slow degradation itself. This situation is also referred to as limited bioavailability. Hence the general objective of this study was to investigate methods to improve bioremediation of PAH contaminated soils, this means to decrease residual concentrations after bioremediation.

In Chapter 2, a literature survey is given of methods that might increase the desorption rate of hydrophobic compounds. Different methods were discussed and evaluated based on their proposed effects. It was found that almost all methods that tried to improve bioremediation by an increase of the pore diffusion rate led to higher initial remediation rates. However, these methods did not lead to lower residual concentrations.
It was found that only a redistribution of contaminants over the soil had proofed to decrease residual concentrations after biodegradation. Redistribution can be performed by soaking of the soil in an organic solvent. Also, an increase of the organic matter diffusivity showed good perspectives for decreasing residual concentrations, although literature data on this were scarce. A diffusivity increase can be performed by a short-term heating of the soil or by an extraction of

multivalent cations from the soil. However the latter is a timely and expensive method. Soaking with an organic solvent and short-term heating were selected for further study.

In Chapter 3, the effects of pretreatment were studied on subsequent biodegradation of PAH contaminants. Short-term heating and soaking with an organic solvent were used as pretreatment methods, as they were selected to be the two most promising methods in the previous chapter. Two different contaminated soils were used for this: a clayey classification sludge with a high organic matter content from a former gas plant site and a sandy soil with a low organic matter content from a wood preservation site. In the classification sludge, heating at 120 °C for one hour increased the percentage of PAH that were removed from 9.5 % to 27 % in a subsequent biodegradation step of 21 days. Lower temperatures resulted in smaller increases of the removal percentages. The observed increase in biodegradation was hypothesised to be caused either by transfer of PAH from sorption sites with low desorption rates to those with high ones or by transformation of slow-desorption sites into fast-desorption sites. Thermal pretreatment of the sandy soil showed no significant effect on the degradation of aged PAH. Soaking of the sludge in a 4:1 ($^{v}/_{v}$) acetone-water mixture, using 0.25 ml solvent per gram of soil, increased the degree of biodegradation from 9.5 % to 20 %, probably as a result of dissolution of the PAH in the pore liquid during soaking. Soaking of the sandy soil increased the biodegradation of only high molecular weight PAH (5 or more aromatic rings), namely from 24 % to 48 %.

In Chapter 4, desorption kinetics of PAH contaminants were studied, because biodegradation of hydrophobic organic contaminants in soil is often limited by slow desorption of these compounds from the soil. It was tried to determine the effects of pretreatment time and pretreatment temperature on desorption kinetics of PAH contaminants. Prior to determining desorption kinetics, the soil was subjected to a temperature increase (60 - 100 °C) for a short period of time (10 min. - 24 h). After heating, desorption kinetics of several PAH were determined using a solid phase extraction technique. Desorption curves could be fitted to a two-compartment model, consisting of a fast and a slow desorbing fraction of contaminants. It was found that the fast desorbing fractions had increased after a thermal treatment. For example, the fast desorbing fraction of pyrene increased from 20 % before thermal treatment to 48 % after a thermal treatment of 24 hr at 100 °C. Lower temperatures and shorter treatment times led to smaller increases of fast desorbing fractions.

Also, in this chapter it was tried to further elucidate the mechanism that causes the increase of the PAH bioavailability as a result of thermal treatment. It was found that the results of this study correspond well with a mechanism in which contaminants that are entrapped within the soil organic matter are released as a result of a thermal pretreatment.

In Chapter 5, desorption kinetics and biodegradation kinetics of PAH contaminants in several soils were compared. It was tried to determine whether desorption experiments (using solid-phase extraction) can predict bioremediation results. Both desorption and biodegradation experiments were performed with soil slurries, which made it possible to directly compare biodegradation and desorption kinetics. Two different soils and one sediment, which were all field-contaminated, were used in the experiments. It was found that not only the fast desorbing PAH could be biodegraded, but also part of the slowly desorbing PAH. Further, biodegradation rates initially controlled removal of both low and high molecular weight PAH compounds. Nevertheless, after 21 days, desorption rates controlled biodegradation of the low molecular weight PAH, and consequently residual concentrations of these compounds were the same for both desorption and biodegradation experiments. This showed that desorption experiments eventually predict biodegradation results for low molecular weight PAH well. On the other hand, for high molecular weight PAH compounds, biodegradation rates were always smaller than desorption rates, which was likely caused by low growth rates of microorganisms that have to degrade these compounds.

In Chapter 6, a mathematical model was developed to describe desorption and biodegradation of PAH and growth of PAH-degrading biomass. Desorption of PAH was described by biphase desorption kinetics incorporating a fast and a slow desorption compartment, as is generally found in PAH desorption. Biodegradation and biomass growth were described using Monod kinetics to which a term for maintenance requirements was added. Results obtained with this model were compared to experimental results of PAH biodegradation and desorption in three different soils. It was found that the model could describe the experimental biodegradation results well. Especially, the model showed that persistence of high molecular weight PAH in soil could be caused by slow growth of biomass that degraded these substances, as had been hypothesised in the previous chapter. This slow growth was attributed to the low aqueous phase concentrations of these high molecular weight PAH, which were too low to meet the maintenance requirements of the microorganisms. Further, the sensitivity of the model was tested for various input parameters. It was found that, particularly,

independent determination of the soil-water partition coefficients of the PAH would further improve the predictive value of the model.

In Chapter 7, the technological and economical possibilities for application of a thermal pretreatment in soil bioremediation were studied. Different techniques for thermal pretreatment in several bioremediation processes for contaminated soils were discussed. Treatment costs of the different pretreatment techniques were calculated using a factorial cost estimate method. The combined costs of thermal pretreatment and subsequent bioremediation were compared to costs of alternative strategies for remediation. It was found that thermal pretreatment can be incorporated successfully in current bioremediation practice for both technical and economical reasons. Especially, the use of steam injection as a thermal pretreatment step in biopile remediation and at in-situ remediation is a competitive alternative to current remediation techniques.

Previous chapters have shown that thermal pretreatments are a potentially successful method to increase the bioavailability of PAH contaminants. In Chapter 8 it has been discussed for what type of bioremediation technologies, soil types, and contaminants a thermal pretreatment can be applied. From this discussion it was concluded that thermal pretreatments could improve results of most bioremediation technologies and most hydrophobic compounds. Also, pretreatments might be best applied for high organic matter soils with no tar constituents.
Further it was discussed whether thermal pretreatment can play an important role in current and future remediation practices. It was found that as long as cleanup guidelines are defined as total contaminant concentrations, thermal pretreatment could be useful in obtaining the guideline concentrations. In case guidelines will be defined as leaching levels, thermal pretreatments will be no longer necessary to reach the guideline levels. For persistent compounds, like high molecular weight PAH, thermal pretreatment might even lead to increased leaching levels.

Samenvatting

Na de ontdekking van grote hoeveelheden chemisch afval onder een woonwijk in Lekkerkerk in 1978, is uit verder onderzoek naar het voorkomen van verontreinigingen gebleken dat op een groot aantal locaties in Nederland de bodem sterk verontreinigd is. Ongeveer één procent van deze verontreinigde grond bevat polycyclische aromatische koolwaterstoffen (PAK). PAK-verontreinigingen staan in de belangstelling, omdat deze stoffen giftige, mutagene en kankerverwekkende eigenschappen hebben. PAK zijn koolwaterstoffen die bestaan uit twee of meer samengevoegde aromatische ringen. Deze stoffen zijn niet vluchtig, slecht oplosbaar in water en hebben de neiging sterk aan de vaste bodembestanddelen te hechten. Dit alles heeft tot gevolg dat verwijdering van PAK uit verontreinigde grond een kostbaar proces is.
Voor de reiniging van grond kunnen zowel fysisch/chemische technieken als biologische methoden worden gebruikt. Biologische reiniging heeft als voordelen boven het gebruik van fysische/chemische technieken dat de bodemstructuur intact blijft, er geen afvalproducten worden gevormd en dat de benodigde energie voor reiniging relatief beperkt is. Desondanks is het vaak niet mogelijk gebleken PAK-verontreinigde gronden op biologische wijze te reinigen als gevolg van trage afbraaksnelheden en te hoge concentraties van verontreinigingen die achterblijven in de bodem. Deze trage en onvolledige afbraak wordt voornamelijk veroorzaakt door een trage desorptie van PAK vanuit de bodemdeeltjes en dus niet vanwege het feit dat de afbraak zelf langzaam verloopt. Dit wordt ook wel omschreven als een beperkte bioschikbaarheid van de verontreinigingen. Derhalve was het algemene doel van dit onderzoek om methodes te ontwikkelen die kunnen leiden tot een verbetering van de biologisch reiniging van PAK-verontreinigde gronden, dat wil zeggen tot lagere restconcentraties na reiniging.

In een Hoofdstuk 2 wordt een literatuuronderzoek gepresenteerd waarin diverse methodes worden besproken die de desorptiesnelheid van PAK en andere slecht oplosbare verbindingen kunnen vergroten en eindconcentraties kunnen verlagen. Uit dit onderzoek bleek dat bijna alle methodes, die poogden de diffusiesnelheid van verontreinigingen door poriën in bodemaggregaten te vergroten, leidden tot

hogere initiële afbraaksnelheden. Deze methodes konden echter geen lagere restconcentraties bewerkstelligen.
Een herverdeling van verontreinigingen door middel van voorweken met een organisch solvent bleek wel te leiden tot lagere restconcentraties. Zo'n herverdeling kan worden uitgevoerd door de grond te weken in een organisch solvent. Verder bood ook het vergroten van de diffusiecoëfficiënt van de verontreinigingen in de organische stof een goede mogelijkheid voor het verlagen van restconcentraties. Zo'n vergroting van de diffusiecoëfficiënt kan mogelijk worden uitgevoerd door grond korte tijd te verhitten of door extractie van multivalente kationen uit de grond. Dit laatste is echter een langdurige en kostbare methode. Voorweken met een organisch solvent en een korte verhitting van de grond werden gekozen voor verder onderzoek.

In Hoofdstuk 3, zijn de gevolgen van voorbehandelingen op de daarop volgende biologische afbraak onderzocht. Een kortdurende verhitting van grond en voorweken met een organisch solvent werden toegepast als voorbehandelingsmethodes, aangezien beide methodes als veelbelovend uit de literatuurstudie naar voren kwamen. Voor dit onderzoek werden twee verschillende gronden gebruikt: een classificatie-slib met een hoog lutum en organisch stof gehalte afkomstig van een voormalig gasfabrieksterrein en een zandgrond met een laag organisch stofgehalte van het terrein van een houtverduurzamingsbedrijf. Bij het classificatie-slib leidde een verhitting van 1 uur bij 120 °C tot een toename van het percentage PAK dat vervolgens in 21 dagen kon worden afgebroken van 9,5 % naar 27 %. Bij lagere voorbehandelings-temperaturen was het effect navenant kleiner. De toename van de biobeschikbaarheid werd verondersteld te zijn veroorzaakt door of een overdracht van PAK naar sorptieplaatsen met lage desorptiesnelheden naar plaatsen met hoge desorptiesnelheden of door een verandering van traag-desorberende in snel-desorberende sorptieplaatsen. Thermische voorbehandeling van de zandgrond leidde niet tot een significante toename van de afbraak van PAK. Voorweken van het classificatieslib met een 4:1 ($^{v}/_{v}$) aceton-water mengsel, met 0,25 ml solvent per gram droge grond, verhoogde het percentage dat afgebroken kon worden van 9,5 % naar 20 %. Dit was waarschijnlijk het gevolg van het oplossen van PAK tijdens het voorweken. Voorweken van de zandgrond verhoogde enkel het gehalte van hoogmoleculaire PAK (5 of meer aromaatringen), namelijk van 24 % naar 48 %.

In Hoofdstuk 4, werd de desorptiekinetiek na (thermische) voorbehandeling bestudeerd, vanwege het feit dat afbraak van slecht oplosbare verbindingen in

grond meestal bepaald wordt door trage desorptie van deze stoffen. De invloed van voorbehandelingstijd en -temperatuur op de desorptiekinetiek van PAK-verontreinigingen werd onderzocht. Alvorens de desorptiekinetiek werd bepaald, werd de grond gedurende korte tijd (10 min. - 24 uur) verhit (60 - 100 °C). Na voorbehandeling werd de desorptiekinetiek bepaald met een zogenaamde 'Solid Phase Extraction'-techniek. De desorptiecurves konden worden beschreven met een model waarbij de PAK-verontreiniging bestond uit een traag en een snel desorberende fractie. Het bleek dat de snel desorberende fractie na voorbehandeling was toegenomen ten koste van de traag desorberende fractie. Bijvoorbeeld, de snel desorberende fractie van pyreen (een PAK met 4 aromaatringen) nam toe van 20 % voorafgaand aan een voorbehandeling tot 40 % na een thermische voorbehandeling van 24 uur bij 100 °C. Lagere temperaturen en kortere verhittingstijden leidden tot navenant kleinere toenames van de snel desorberende fracties.
Verder werd in dit hoofstuk getracht het mechanisme dat ten grondslag ligt aan deze toename van de biobeschikbaarheid meer te verduidelijken. De resultaten van dit onderzoek bleken goed overeen te komen met een mechanisme waarbij verontreinigingen die ingesloten zitten in de bodemorganische stof vrijkomen ten gevolge van een thermische behandeling.

Hoofdstuk 5 beschrijft een vergelijking tussen de desorptiesnelheden en afbraaksnelheden van PAK-verontreinigingen in diverse gronden. Gebruik makend van een 'Solid Phase Extraction'-techiek werd getracht te bepalen of desorptie-experimenten biologische afbraak kunnen voorspellen. Zowel desorptie- als afbraakexperimenten werden uitgevoerd onder slurry-omstandigheden, zodat een directe vergelijking tussen afbraak- en desorptiekinetiek mogelijk was. Twee verschillende gronden en een sediment, welke alle drie praktijk-verontreinigd waren, zijn gebruikt voor de experimenten. Uit de resultaten van de experimenten bleek dat niet alleen de snel desorberende PAK werden afgebroken, maar ook een deel van de traag desorberende PAK. Verder bleek dat de initiële verwijderingssnelheid van de PAK bepaald werd door de afbraaksnelheid. Niettemin bepaalde na 21 dagen de desorptiesnelheid de verwijderingssnelheid van laag moleculaire PAK. Dit had tot gevolg dat uiteindelijke restconcentraties van desorptie- en biodegradatie-experimenten overeenkwamen. Dit betekent dat desorptie-experimenten gebruikt kunnen worden om de resultaten van biologische afbraak te voorspellen. Voor hoog moleculaire PAK waren afbraaksnelheden echter altijd lager dan de desorptiesnelheden, hetgeen hoogstwaarschijnlijk werd veroorzaakt door te lage groeisnelheden van de micro-organismen die deze PAK moeten afbreken.

In Hoofdstuk 6 is met behulp van model getracht de desorptie en afbraak van PAK en groei van PAK-afbrekende micro-organismen te beschrijven. De desorptie van PAK werd beschreven met een twee-fasen model, bestaande uit een snel- en een traag-desorberende PAK-fractie. Deze twee-fasen desorptie is kenmerkend voor PAK-verontreinigingen. Afbraak en groei van biomassa zijn beschreven middels Monod-kinetiek aan welke een term voor onderhoud van biomassa is toegevoegd. De resultaten van dit model werden vergeleken met experimentele resultaten van afbraak en desorptie van PAK in drie verschillende gronden. Het bleek dat het model de experimentele resultaten goed kon beschrijven. Het model toonde met name aan dat slechte afbraak van hoog moleculaire PAK in bodem veroorzaakt kan worden door trage groei van de biomassa die deze verbindingen moeten afbreken. Deze trage groei wordt veroorzaakt door de zeer lage concentraties in de waterfase van deze hoog moleculaire PAK; deze concentraties zijn te laag om in de onderhoudsbehoeften van de micro-organismen te voorzien. Verder is de gevoeligheid van het model bepaald voor de diverse invoerparameters. Het bleek dat de voorspellende waarde van het model verbeterd kon worden, door een onafhankelijke bepaling van de grond-water-verdelingscoëfficiënt van de PAK.

In Hoofdstuk 7 zijn de technische en economische mogelijkheden voor toepassing van thermische behandelingen bij biologische bodemreiniging onderzocht. Verschillende technieken voor een thermische voorbehandeling en verschillende biologische reinigingsprocessen zijn besproken. De kosten van de verschillende voorbehandelingstechnieken zijn berekend met behulp van een factoriele kostenschatting. De kosten van een gecombineerde voorbehandeling en biologische reiniging zijn vergeleken met alternatieve reinigingstechnieken. Het bleek dat een thermische voorbehandeling goed kan worden geïmplementeerd in de bestaande biologische reinigingspraktijk, zowel op technische als op economische gronden. Met name het gebruik van stoominjectie als thermische voorbehandelingsstap bij landfarming en in-situ reiniging is een concurrerend alternatief voor bestaande reinigingstechnieken.

De vorige hoofdstukken lieten zien dat een thermische voorbehandeling een succesvolle methode kan zijn om de biobeschikbaarheid van PAK-verontreinigingen te verhogen. In Hoofdstuk 8 wordt besproken voor welke biologische reinigingstechnieken, voor welke grondsoorten en voor welke verontreinigingen een thermische voorbehandeling toegepast kan worden. Uit deze discussie werd geconcludeerd dat een thermische voorbehandeling de

reinigingsresultaten van de meeste technieken en de meeste hydrofobe verbindingen kan verbeteren. Daarnaast kan een thermische voorbehandeling waarschijnlijk het beste toegepast worden bij grond met een hoog organisch stofgehalte en waarin geen teerdeeltjes aanwezig zijn.
Verder werd besproken wat het belang kan zijn van een thermische voorbehandeling binnen de huidige en toekomstige bodemreinigingspraktijk. Het bleek dat zo lang als reinigingsnormen bepaald worden door eindconcentraties na reiniging, een thermisch voorbehandeling zeer bruikbaar kan zijn bij het behalen van deze normen. Indien echter, in plaats van concentraties, uitloogniveaus als norm gebruikt zullen worden, zijn voorbehandelingen niet langer noodzakelijk. Voor moeilijk afbreekbare verbindingen zoals hoog moleculaire PAK, kan een thermische voorbehandeling zelfs leiden tot verhoogde uitloogniveaus.

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Luc Bonten,
Wageningen, 28 november 2000.

Curriculum Vitae

Luc Theodorus Christiaan Bonten werd geboren op 25 augustus 1971 te Roermond. Na behalen van het Gymnasium-β diploma aan het Bischoppelijk College Broekhin te Roermond in 1989, begon hij met een studie Chemische Technologie aan de Universiteit Twente te Enschede. In eerste instantie stortte hij zich op de 'echte' procestechnologie, uitmondend in een stage bij de afdeling Industriële Katalyse van DSM-Research te Geleen. Hierna heeft hij zich echter bekeerd tot de milieutechnologie en in 1994 zijn studie afgerond met een afstudeeronderzoek naar de biologische afbraak van BTEX-componenten in industriëel afvalwater bij de onderzoeksgroep Technisch Milieubeheer. Korte tijd daarna werd hij aangesteld als assistant in opleiding bij de vakgroep Milieutechnologie van de Landbouwuniversiteit Wageningen. Het betrof een onderzoek naar de verbetering van de biologische afbraak van PAK-verontreinigingen in grond. De resultaten van dit onderzoek zijn weergegeven in dit proefschrift. Sinds oktober 1999 is hij werkzaam als docent proces- en milieutechnologie bij de opleiding Milieukunde van de Hogeschool Brabant te Breda.