January 2010

Impact Of Aging Time On The Risk From Dermal Exposure To Soil Contaminated With Phenanthrene

Mohamed S. Abdel-Rahman
*University of Medicine and Dentistry of New Jersey*

Gloria A. Skowronski
*University of Medicine and Dentistry of New Jersey*

Rita M. Turkall
*Clinical Laboratory Sciences Department*

Follow this and additional works at: [https://scholarworks.umass.edu/soilsproceedings](https://scholarworks.umass.edu/soilsproceedings)

**Recommended Citation**
PART X: Risk Assessment

Chapter 30

IMPACT OF AGING TIME ON THE RISK FROM DERMAL EXPOSURE TO SOIL CONTAMINATED WITH PHENANTHRENE

Soil-aged Phenanthrene Dermal Exposure Risk

Mohamed S. Abdel-Rahman¹, Gloria A. Skowronski¹, and Rita M. Turkall¹,²

University of Medicine and Dentistry of New Jersey,¹Pharmacology and Physiology Department, New Jersey Medical School and ²Clinical Laboratory Sciences Department, School of Health Related Professions

Abstract: The health risk from exposure to contaminated soil is related to the fraction of chemical absorbed by the body (bioavailability), rather than to the total concentration of chemical in soil. Chemical bioavailability data are necessary to improve the accuracy of risk assessment following exposure to contaminated soil and to allow more realistic soil remediation goals. One of the factors that may influence chemical bioavailability and ultimately health risk from exposure is the residence time or “aging” of chemical in soil. Skin is a primary route of exposure to phenanthrene, a polycyclic aromatic hydrocarbon found in soil at former manufactured gas plant sites. This study was conducted to determine the extent to which soil alters the dermal bioavailability of phenanthrene with respect to soil aging and soil type. Bioavailability was assessed by measuring the penetration of phenanthrene through dermatomed male pig skin via an in vitro approach consisting of radiotracer and flow-through diffusion cell methodology. After 3 months aging, dermal penetration was significantly decreased by 83% in Atsion soil (high sand and high organic matter content) and by 69% in Keyport soil (high clay but low organic matter content) versus pure phenanthrene (without soil). By extending the aging time to 6 months, penetration through skin was reduced by 94% in Atsion soil and 86% in Keyport soil. The results indicate that because human risk from exposure to soil contaminated with phenanthrene would be reduced by aging, less soil cleanup would be needed.

Key words: bioavailability, polycyclic aromatic hydrocarbon, skin, matrix effects

1. INTRODUCTION

Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous contaminants of soil and are derived chiefly from the incomplete combustion of organic materials. Soils that contain waste from manufactured gas plant sites, petroleum refineries, and wood preserving facilities are among the most heavily contaminated with PAHs (Harvey, 1991; ATSDR, 1995; Loehr and Webster, 1997). The fate of PAHs in soil has caused increasing concern due to their toxic, mutagenic, and carcinogenic effects (Karimi-Lotfabed et al., 1996). Health risk assessments are often based on exposure to the total concentration of a chemical in a contaminated site. The total concentration is usually determined by rigorous extraction procedures such as acid digestion, sonication, or Soxhlet extraction to remove the chemicals from soil (USEPA, 1986, 1992; Tang et al., 1999). However, this approach can result not only in an overestimation of risk, if the total amount of a chemical is not available to the body, but also in unnecessary and costly soil cleanup.
When humans are exposed to a chemical in soil, only a fraction of the total concentration (the bioavailable fraction) may be absorbed into the systemic circulation and reach the target site. It is the bioavailable amount that determines if chemical exposure produces a toxic effect or the extent of that effect. Therefore, it is important to determine the bioavailability of a chemical in soil to improve the accuracy of risk assessment following exposure to contaminated soil affording more realistic assessments of health effects and of soil remediation goals.

A number of factors can influence chemical sorption to soil and subsequently affect bioavailability. These factors include soil properties (e.g. percent organic matter, clay content, pH, and particle size) (Karickhoff et al., 1979; Duffus, 1980; NEPI, 2000; Pu et al., 2004); chemical properties (such as lipophilicity and volatility) (Ibbotson et al., 1989), as well as chemical aging in soil (Alexander, 2000).

The movement of chemicals from the surface of soil particles into less accessible sites where they become sequestered over a period of time has been termed “aging” (Linz and Nakles, 1997; Reid et al., 2000). It is believed that during chemical aging, hydrophobic molecules move slowly into soil organic matter. Because soil organic matter is very heterogeneous and contains both nanopores and regions that vary in polarity, density, and degree of coil (Aochi and Farmer, 1997), it has been proposed that hydrophobic molecules can become sequestered in the nanopores as well as the solid phase of the organic matter so that they are no longer readily accessible (Alexander, 2000). Chemical aging in soil has been shown for organic compounds (Steinberg et al., 1987; Bowmer, 1991; Scribner et al., 1992; Hatzinger and Alexander, 1995; Kelsey et al., 1997; Robertson and Alexander, 1998; Roy and Singh, 2001; Abdel-Rahman et al., 2002, 2004, 2006) as well as for heavy metals (Lock and Jannsen, 2003; Turpeinen et al., 2003; Abdel-Rahman et al., 2005).

The present study focused on the effects of aging on the dermal bioavailability of phenanthrene in soil. Phenanthrene is a major polycyclic aromatic hydrocarbon emitted in coal and fossil fuel combustion. It is found in coal combustion wastes such as coal tar, which has contaminated soils globally at sites where gas was formally manufactured (Laor et al., 1999; Pu et al., 2004). With the advent of natural gas pipelines, the gas manufacturing plants were shut down and the tars were generally left on site or buried in landfills (Wyzga and Goldstein, 1994). The composition of various coal tars depends on the source of the coal tar and the methods of processing. For example, when Chu et al. (1988) investigated the subchronic dermal toxicity of a medium-boiling (bp 154-378°C) coal liquefaction product in the rat, they found that the predominant polycyclic aromatic hydrocarbon was phenanthrene, comprising 27% w/w of the total chemical composition. Smaller amounts of fluorene, fluoranthrene, acenaphthene, 2-methylanthracene, pyrene, and 9,10-dihydrophenanthrene were also present. There are 868 hazardous waste sites contaminated with phenanthrene in the United States (ATSDR, 2006). Soil concentrations of phenanthrene at contaminated sites can range from 150 - 716 mg/kg dry weight at former manufactured gas plant sites to 11 - 4,434 mg/kg at sites related to wood preservation processes (Wilson and Jones, 1993).

Phenanthrene can be inhaled from exposure to coal, wood, and cigarette smoke; and from gasoline and diesel engine exhaust. Phenanthrene is also present in charcoal-broiled food. However, the most likely route of exposure to phenanthrene in contaminated soil is via the dermal route. Other sources of dermal exposure to phenanthrene include waste water, used motor oil, crude oils, and lubricating oils (IARC, 1983; ATSDR, 1995).

Acute health effects may occur immediately or shortly after dermal exposure to phenanthrene. The skin can become irritated. If skin contaminated with phenanthrene is exposed to sunlight, a rash or skin burn may occur, sometimes with blisters. Chronic health effects can occur some time after exposure to phenanthrene and can last for months or years. If a skin allergy develops, very low future exposure can cause itching and a skin rash (NJDHSS, 1999).

The aim of this study was to assess the bioavailability of phenanthrene in soil by measuring the dermal penetration of the chemical. First, the dermal penetration of phenanthrene in freshly spiked soil was compared to the dermal penetration of pure phenanthrene (without soil). Although bioavailability data for freshly spiked soil can overestimate the dermal penetration of a chemical that has been in the same soil for a longer period of time, the data on freshly spiked soil are important because they can be used to predict the risk from newly contaminated soil. However, bioavailability...
can be decreased more by aging in soil than when the soil is newly contaminated. Therefore, the effects of aging in soil on further reducing the dermal penetration of phenanthrene were investigated. Because soil type (sand, clay) and the percent of organic matter in soil are factors that will influence chemical sorption, their effects on dermal penetration were also examined. From the data generated by this study, it was possible to evaluate the impact of chemical aging on the health risk from phenanthrene exposure. Environmentally acceptable endpoints (EAEs) can then be established for phenanthrene that has been aged in soil. The EAE is the concentration of chemical in soil that will not adversely affect human health and the environment (Alexander, 1995).

2. MATERIALS AND METHODS

2.1 Chemicals

Phenanthrene (9,10-14C), in ethanol, with a specific activity of 12.8 mCi/mmole and a radiochemical purity of 99% was purchased from New England Nuclear (NEN) Life Science Products, Boston, MA. Prior to use, the radioisotope was diluted with U.S.I. pure ethyl alcohol, dehydrated U.S.P. (U.S. Industrial Chemicals Co., Division of National Distillers and Chemical Corp., NY).

2.2 Soils

Studies were performed on two different soils. Both are representative of soil types widely distributed in the United States. The Atson soil consists of 90% sand, 8% silt, 2% clay, 4.4% organic matter; has a pH of 4.2; and was collected from the Cohanekey sand formation near Chatsworth in south central New Jersey (USDA, 1977). The Keyport soil contains 50% sand, 28% silt, 22% clay, 1.6% organic matter; has a pH of 5.0; and was collected from the Woodbury formation near Moorestown in southwestern New Jersey (USDA, 1972). The majority of the soil particles were 50-250 µm in size. Soil analyses were performed by the Soil Testing Laboratory at Rutgers Cooperative Extension Resource Center, Rutgers University, New Brunswick, NJ. Organic matter content was measured by a modified Walkley and Black (1934) dichromate oxidation method.

2.3 Chemical Aging in Soil

Radiolabeled phenanthrene was added to each of the soils that had been previously autoclaved and hydrated to 11% (w/w) with sterile distilled-deionized water. This is the maximum amount of water that could be used to lightly moisten the soils without there being an excess of water when phenanthrene was added to the soils. Soil moisture content is an important factor in contaminant sorption (Unger et al., 1996) because water may compete with organic molecules for adsorption sites on soil (Ruiz et al., 1998). Phenanthrene was added to soil at a ratio of 100 µg to 1 g of soil. After phenanthrene was mixed thoroughly with the soils to ensure uniform distribution of chemical, spiked soils were added to Teflon-sealed vials and stored in the dark at room temperature for 3 months and 6 months.

2.4 Animal Model

Whole pig skin was obtained from the costo-abdominal areas of euthanized (40-60 lb) male Yorkshire pigs (Cook College Farm, Rutgers University, New Brunswick, NJ). The pig has been widely accepted as an animal model for studying human percutaneous absorption of a large variety of chemicals under various experimental conditions (Bartek et al., 1972; Reifenrath and Hawkins, 1986; Qiao et al., 1993) because of the well documented histological (Monteiro-Riviere and Stromberg, 1985), physiological, biochemical, and pharmacological similarities between pig skin and human skin.
(Qiao and Riviere, 2000). Skin was transported to the laboratory and viability maintained in ice-cold HEPES buffered (25 mM) Hank’s balanced salt solution (HBBSS), pH 7.4, containing gentamycin sulfate (50 mg/l) (Collier et al., 1989) after which it was immediately prepared for diffusion cells according to Bronaugh and Stewart (1985).

2.5 In Vitro Dermal Penetration Studies

Excised skin was cut to a thickness of 200 µm with a dermatome (Padgett Electro-Dermatome Model B, Padgett Instruments Inc., Kansas City, MO) and circular pieces were mounted into Teflon flow-through diffusion cells (Crown Bio Scientific, Inc., Somerville, NJ). The exposed skin surface area (0.64 cm²) was maintained at a temperature of 32°C. The dermal side of each skin sample was perfused with HBBSS containing 10% fetal bovine serum (Sigma/Aldrich, St. Louis, MO) at a flow rate of 3 ml/h and aerated continuously with oxygen (Collier et al., 1989). Chemical was applied to the surface of the skin either alone in 5 µl of ethanol vehicle, immediately after the addition of 30 mg of soil, or after aging in 30 mg of each of the two soils. The chemical dose was 5.2 µg/cm² skin. Receptor fluid (perfusate) was collected in scintillation vials containing 10 ml of Formula-989 liquid scintillation cocktail (Packard Instruments Co., Inc., Meriden, CT) up to 24 h postdosing. Loosely adsorbed chemical was washed from the skin surface with soap and water (once with 1 ml of a 1% aqueous soap solution and twice with 1 ml of distilled-deionized water). Skin samples were completely solubilized in Solvable (Packard) for 8 h at 50°C to determine the quantity of the chemical remaining in skin. Radioactivity in all samples was counted by liquid scintillation spectrometry (LS 7500, Beckman Instruments, Inc., Fullerton, CA). Sample quench was corrected by using the H-ratio method.

2.6 Statistical Analysis

All data were reported as the mean ± standard error of the mean (SEM) and expressed as percent of the initial dose. Statistical differences between treatment groups were determined by one-way analysis of variance (ANOVA) with Scheffe’s test except for the soil comparisons which were performed by Student’s independent t-test. The level of significance was p < 0.05.

3. RESULTS

A comparison between the dermal penetration of pure phenanthrene and phenanthrene in the Atsion soil (as freshly spiked or aged) is shown in Table 1. When soil was freshly spiked, the dermal penetration of phenanthrene was significantly decreased versus pure chemical. Four percent of the pure phenanthrene dose penetrated into the receptor fluid during the 24 h collection period. However, only one-half that amount was detected for phenanthrene in the freshly spiked Atsion soil. Lipophilic compounds have a tendency to form reservoirs in skin (Chu et al., 1996). Therefore, when dermal penetration studies are conducted, it is also necessary to determine the amount of chemical remaining in skin because chemical that forms a reservoir in skin has the potential to be absorbed systemically with time. In this study, most of the pure phenanthrene dose (61%) was detected in skin. Atsion soil freshly spiked with phenanthrene reduced the amount of phenanthrene in skin to 24% of the initial dose. The total penetration represents the sum of the chemical penetrating into the receptor fluid and the amount in skin (Chu et al., 1996). Because skin contained more radioactivity than receptor fluid, the percent of the initial phenanthrene dose comprising total penetration was very similar to that in skin (65% for pure and 26% for freshly spiked soil).

<table>
<thead>
<tr>
<th>Table 1. Effect of Aging Time in Atsion Soil on the Dermal Penetration of Phenanthrene</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure</td>
</tr>
<tr>
<td>------</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>

https://scholarworks.umass.edu/soilsproceedings/vol12/iss1/31
Further reductions in the dermal penetration of phenanthrene into receptor fluid and skin were observed after aging in Atsion soil. Only 0.4% and 0.3% of the initial dose, respectively, were detected in receptor fluid after 3 and 6 months aging. Likewise, radioactivity in skin was decreased to 11% and 4%, respectively. Consequently, total penetration was lowered to 11% of the initial dose and to 4% after 6 months. The skin wash data correlated well with the results for total penetration. Only 6% of the radioactivity applied to skin was found in the skin wash for the pure compound. As less phenanthrene penetrated skin, more radioactivity remained on skin (73-96%).

For freshly spiked Keyport soil, penetration into receptor fluid was reduced to about one-third of phenanthrene without soil (Table 2). Skin contained 16% of the dose while total penetration was 17%. Although the total penetration of phenanthrene aged for 3 months in Keyport soil (20%) was similar to that in the freshly spiked soil, results revealed a further decrease to 9% of the dose after 6 months of aging. An increase in the amount of radioactivity retained by soil was also evident in the skin wash with aging.

### Table 2. Effect of Aging Time in Keyport Soil on the Dermal Penetration of Phenanthrene

<table>
<thead>
<tr>
<th></th>
<th>Pure</th>
<th>Freshly Spiked</th>
<th>Aged 3 Months</th>
<th>Aged 6 Months</th>
</tr>
</thead>
<tbody>
<tr>
<td>Receptor Fluid</td>
<td>4.2 ± 0.6 a</td>
<td>2.1 ± 0.3 b</td>
<td>0.38 ± 0.03 b</td>
<td>0.26 ± 0.03 b</td>
</tr>
<tr>
<td>Skin</td>
<td>61.2 ± 3.7</td>
<td>23.8 ± 1.5 b</td>
<td>10.7 ± 1.6 b</td>
<td>3.8 ± 0.5 b</td>
</tr>
<tr>
<td>Total Penetrat.</td>
<td>65.4 ± 3.9</td>
<td>25.9 ± 1.4 b</td>
<td>11.1 ± 1.7 b</td>
<td>4.0 ± 0.6 b</td>
</tr>
<tr>
<td>Skin Wash</td>
<td>5.9 ± 0.6</td>
<td>73.1 ± 1.1 b</td>
<td>88.9 ± 1.7 b</td>
<td>96.0 ± 0.6 b</td>
</tr>
</tbody>
</table>

- a Mean ± SEM of percent initial dose for n = 10-11 replicates per treatment from 3 pigs
- b Significantly different from pure (p < 0.05, ANOVA)
- c Significantly different from freshly spiked (p < 0.05, ANOVA)

When the total penetration of phenanthrene was compared between the two soils (Table 3), it was found that the total dermal penetration of phenanthrene in freshly spiked Keyport soil (17%) was significantly lower than that in the Atsion soil (26%). However, the total dermal penetration of phenanthrene in the Atsion soil was about one-half that in the Keyport soil for both aging times.

### Table 3. Effect of Soil Type on the Dermal Penetration of Phenanthrene

<table>
<thead>
<tr>
<th>Time in Soil</th>
<th>Atsion Soil</th>
<th>Keyport Soil</th>
</tr>
</thead>
<tbody>
<tr>
<td>Freshly Spiked</td>
<td>25.9 ± 1.4 a</td>
<td>17.4 ± 2.5 b</td>
</tr>
<tr>
<td>3 Months</td>
<td>11.1 ± 1.7</td>
<td>20.5 ± 3.5 b</td>
</tr>
<tr>
<td>6 Months</td>
<td>4.0 ± 0.6</td>
<td>9.1 ± 2.1 b</td>
</tr>
</tbody>
</table>

- a Percent total penetration (mean ± SEM) for n = 8-11 replicates per treatment from 3 pigs
- b Significantly different from the Atsion soil (p < 0.05, Student’s independent t-test)

### Discussion and Conclusions

Dermal exposure to pure phenanthrene is a potential health hazard because a reservoir of chemical was detected in skin. The formed reservoir may contribute to skin sensitization which may eventually cause skin rashes and blisters. However, a decrease in dermal penetration was observed when phenanthrene was sorbed to a soil matrix. The dermal penetration of phenanthrene was significantly decreased by 60% and 73%, respectively, in freshly contaminated Atsion and Keyport soils relative to pure chemical.

It is well established that soil organic matter acts primarily as a partition medium, while mineral matter acts as an adsorbent in the sorption of nonionic organic compounds in soil (Karickhoff et al.,...
In this study, a lower total penetration of phenanthrene as well as a higher percentage of clay in the Keyport soil (11 times greater than Atsion soil) suggests that more surface adsorption occurred in that soil when it was freshly spiked.

After 3 months of aging, the dermal penetration of phenanthrene was reduced by 83% in the Atsion soil. However, for the Keyport soil, 3 months aging did not reduce penetration through skin more than the newly contaminated soil. Instead, a longer aging time was needed to decrease the dermal penetration of phenanthrene further in that soil. By extending the aging time to 6 months, lowered penetration through skin was reduced by 94% in the Atsion soil and by 86% in the Keyport soil. Also, the dermal penetration of phenanthrene was about 50% less when aged in the Atsion soil than in the Keyport soil. The Atsion soil contains three times as much organic matter as the Keyport soil which indicates that with time, more sequestration into organic matter occurred in that soil. Nam et al. (1998) suggested that the organic matter content of soil is a major determinant of sequestration. In their studies, sequestration was measured by the extent of mineralization of phenanthrene by an added bacterium. Furthermore, the data of Nam and Kim (2002) suggested that major sequestration sites for aged phenanthrene may reside in the humin-mineral fraction of soil.

Since the effects of phenanthrene on human health are dependent on the dermal bioavailability of the chemical, the data indicate that the potential health risks from dermal exposure to phenanthrene would be reduced by soil and aging compared to phenanthrene without soil. The data from this study can have an impact on the EAEs of phenanthrene. EAEs were calculated by dividing the total penetration of pure phenanthrene by the total penetration of phenanthrene aged in soil for 3 or 6 months. Three months aging in the Atsion soil raises the EAE for phenanthrene 6-fold compared to the chemical without soil. After aging for 6 months, the EAE for phenanthrene increases 18-fold. In the Keyport soil, the increase in the EAE for 3 months aged phenanthrene would be 3.4-fold compared to the pure chemical while 6 months aging in the same soil increases the EAE to 11.7-fold versus the chemical alone. With increased EAEs, less soil cleanup would be required at contaminated sites where phenanthrene aging has occurred. As a result, soil remediation costs would be lower and there would be fewer site restrictions.

REFERENCES

Impact of Aging Time on the Risk From Dermal Exposure to Soil Contaminated with Phenanthrene

Abdel-Rahman et al.: Dermal Exposure To Soil Contaminated With Phenanthrene


