

## OPTIMIZATION OF SOIL CONDITIONS FOR BIOREMEDIATION IN CHEMICALLY PRETREATED SOILS: SITE CHARACTERIZATION

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### Abstract

Recent focus of remediation of contaminated subsurface and groundwater with organic compounds has shifted towards *in situ* based strategies, such as *in situ* bioremediation and *in situ* chemical oxidation (ISCO). The coupling of these two techniques into an efficient and effective technology requires further investigation into the optimization of bioremediation in chemically pretreated soil. To this end, a sampling trip to Węglińiec, Poland, a site within the EU consortium Upsoil, was planned and executed, as described here. Sampling locations at the site were chosen by integrating previous site investigations with the experimental setup and scientific goals of this project. By taking into consideration the influence of the type of field material on the relevancy of the experimental results, sampling was performed to ensure that findings from this project will be general conclusions transferable to a variety of sites requiring remediation.

**Keywords:** site characterization, sampling strategy, subsurface contamination, remediation, *in situ* chemical oxidation, bioremediation

### Introduction

Subsurface and groundwater contamination of xenobiotic organic compounds has created a large international market for remediation of these polluted sites valued at US\$ 50-60 billion [1]. Conventional *ex situ* and on site technologies are being replaced by *in situ* techniques, where degradation of contaminants is contained within the subsurface [2]. *In situ* bioremediation and, more recently, *in situ* chemical oxidation (ISCO) are arguably the most commonly used *in situ* treatments, which provide rapid and cost effective remediation under a variety of subsurface conditions in combination with various contaminant types.

ISCO is a remediation technology designed to rapidly remove pure product within the source zone area of a polluted site [3, 4]. In the course of a treatment, oxidants such as hydrogen peroxide (Fenton's Reagent), permanganate, persulfate or ozone, are pumped into the subsurface to chemically remove contaminants. Chemical oxidation of a variety of contaminants has been observed, including removal of petroleum derived hydrocarbons [5, 6], polyaromatic hydrocarbons (PAH) [7-9], and chlorinated hydrocarbons [10-12]. Although ISCO is a versatile technology, chemical oxidation is ill-suited for areas outside of the source area. In situations in which the pollutant is dispersed, oxidation of soil organic matter consumes a larger fraction of chemicals than when pure product is present. Rather, bioremediation, although a lengthier overall process, is more appropriate for the plume. As opposed to the source area, lower contaminant concentrations in the plume do not impede biodegradation [1]. Both natural attenuation as well as biostimulation with electron acceptors or nutrients are effective remediation strategies for a variety of contaminant types [13-18].

The combination of chemical oxidation with bioremediation could provide an effective cleanup strategy able to remediate both the source and plume areas of a contaminated site. Although the oxidative stress associated with ISCO treatments is initially detrimental to the microbial population [19-21], work indicates that bacterial populations do regenerate degradation ability [22-24]. In many cases, chemical pretreatment improves bioremediation through decreasing contaminant concentrations to less toxic levels [25] and increasing biodegradability

and bioavailability of the compound and oxidized daughter products [8, 9, 19, 26].

With the aim of ascertaining the feasibility of coupled chemical and biological treatment, field samples from a well characterized contaminated site are required for laboratory experiments. Natural material more closely represents actual site conditions, as (1) pollutants have undergone normal weathering and sorption processes, (2) pure product is present in equilibrium with the soil matrix, and (3) endogenous microbial populations are present. In order to understand experimental results and to facilitate the transfer of conclusions to other sites, an extensive characterization of the contaminated location is required. This includes both background on the history of use and course of pollution at the site, as well as a thorough (hydro)geochemical description of the soil lithology and contaminant distribution. Although normal characterization of a site includes the collection of groundwater and, in some cases, soil samples, additional attention is required when the material will be used for experiments from which more generic conclusions on remediation strategies will be drawn. In this study, we describe how background information on a contaminated site was used to create a sampling strategy and describe the material collected.

#### **Site summary:**

The Polish site Węgliniec was identified within the EU consortium Upsoil as an available location for the development of and experiments with sustainable remediation technologies. It is a railway refueling station which was contaminated with diesel from 1970-2000. In 1997 and in 2001, monitoring wells were sunk over the upper 5-8 meters to both sample the soil TPH concentrations and monitor the groundwater (Figure 1). During a previous remediation project in 2003, an additional 89 wells of 5 meters deep were sunk in a tight grid like pattern. These wells are joined with submerged pipes lying around 30 cm below the ground level. The upper 5 meters were pumped and skimmed to remove free phase oil. After 4 months of pumping it was determined that this remediation project was not successful due to clogging of the filters and was aborted.

The initial location of refueling and thus contamination is near monitoring wells B/01 and B/02, at the southwest corner of the site (Figure 1). In this area, up to 70 cm of pure product is observed in piezometers, and solid phase concentrations are higher than 20 000 mg/kg dry matter. These high pure product concentrations are found at a depth of 150-210 cm below the surface at the same level as the groundwater. Pure product contamination has spread across the site in the same direction as the groundwater, which is in a north-northeast direction. This contamination is also observed on the far side of the site in the brook. This brook is fed by a pipe carrying groundwater through the site. The extent of weathering varies across the site, as concluded from the color and texture of the contamination observed in piezometers. Very low concentrations of BTX are found, indicating that either especially heavy diesel was used or that BTX has already been broken down.

The Węgliniec site was originally the location of a brook running from the south to the north of the site (Figure 2). This created a base peat and clay layer on top of sand (Figures 3 and 4). To the east of the brook, higher sandy dunes were originally present (Figure 2). In order to level the site for the creation of the railroad, anthropogenic fill was added on top of the peat/clay layers (Figures 3 and 4). The composition of the fill is heterogeneous, containing coarse sand, gravel, and somewhat brown finer sand. Thus, at the contaminated site itself, there are 3-4 layers: anthropogenic fill, followed by peat and/or clay, and finally natural sand. In the source area, TPH contamination, which coincides with the groundwater level, occurs within the peat and/or clay layer. Farther to the north, the anthropogenic fill is thicker, thus the contamination is found within this fill layer. To the east of the site, essentially 2 layers are present, an upper more organic rich brown sand followed by sand that is very light in color.

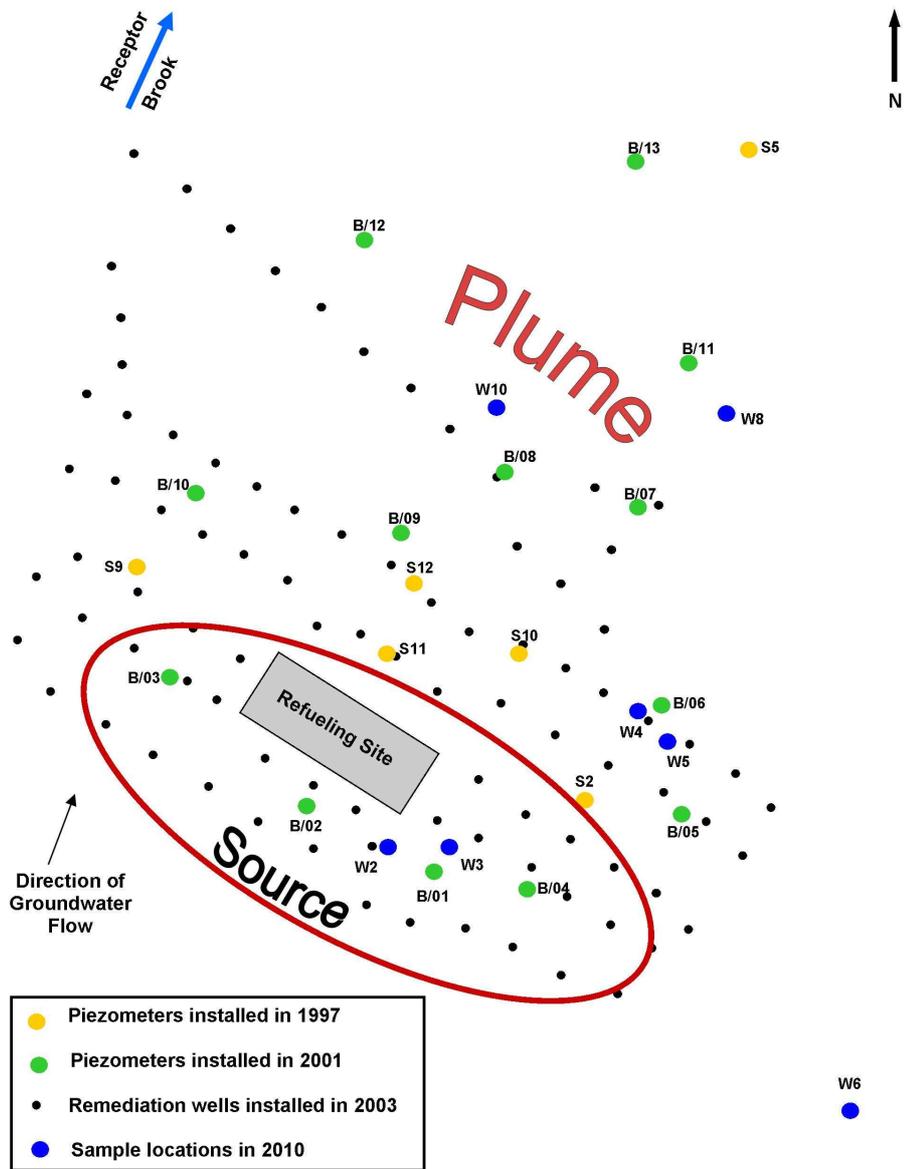


Figure 1. Węgliniec, Poland contaminated railway site, including existing piezometers, remediation wells, and sampling locations in the center of the site.

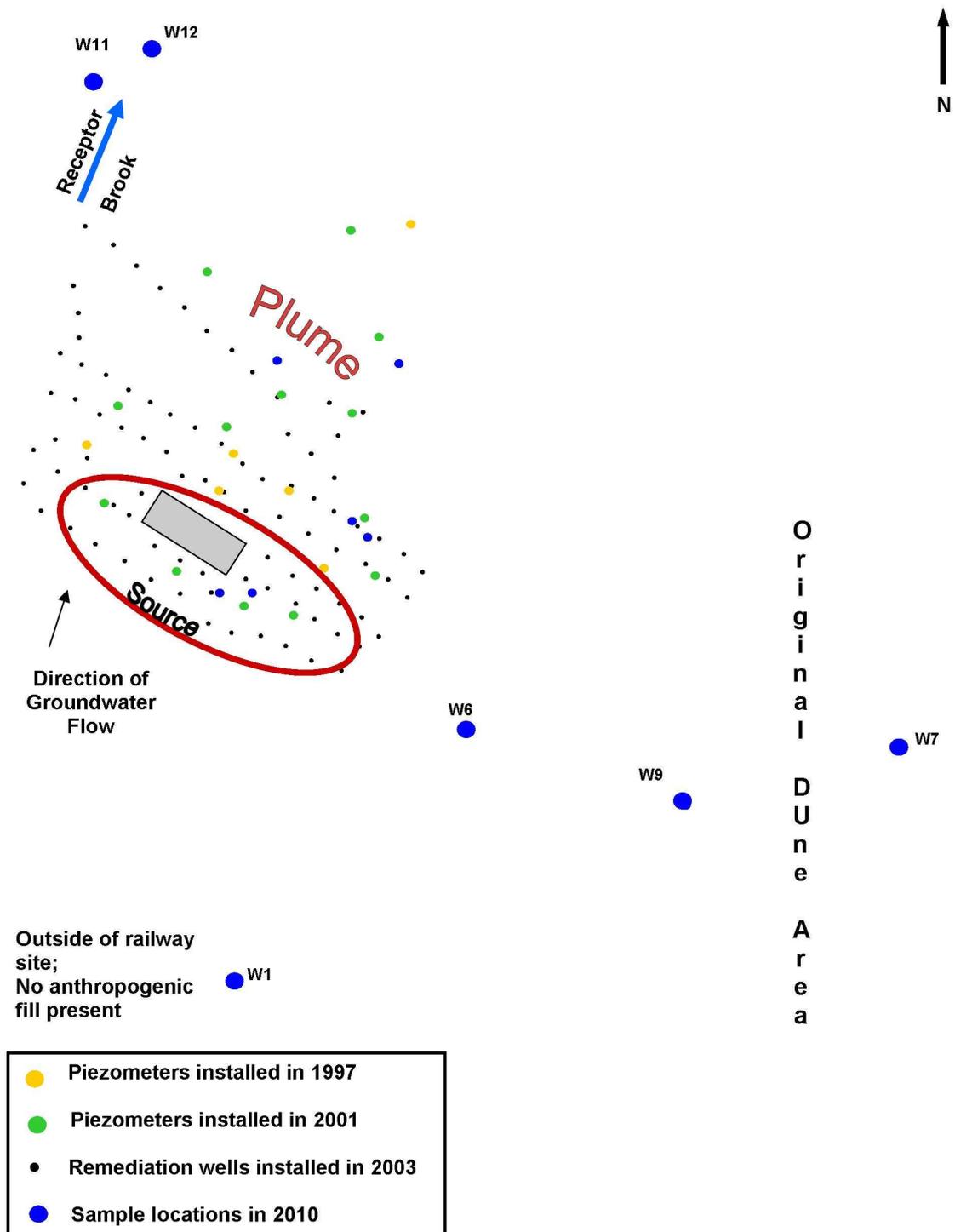


Figure 2. General overview of Węgliń, Poland contaminated railway site which shows the relationship between existing piezometers and the outermost sampling locations.

### Materials and Methods

**Soil Sample Collection:** The exact point of sampling locations as well as the position of monitoring wells was measured using GPS coordinates. At each location, the first 10-40 cm of soil was dug away with a shovel. Then, hand augers with various end shapes were used to remove layers of sediment. These were spread across plastic bags in order to obtain a visual impression of the lithology at the specific location. Samples were collected at various depth ranges (Table 1). Large HDPE containers of 10, 15 or 26 L were taken of soil samples. Pure product was collected from soil samples or from piezometers in 250 mL glass jars. All samples were stored at 4°C upon arrival in The Netherlands.

From the material collected in large containers, sub samples were collected for molecular analysis in sterile 50 mL tubes using a metal spatula. These samples were immediately frozen in liquid nitrogen until bubbling subsided. Samples were stored on dry ice for transport to The Netherlands and were stored at -80°C for analysis.

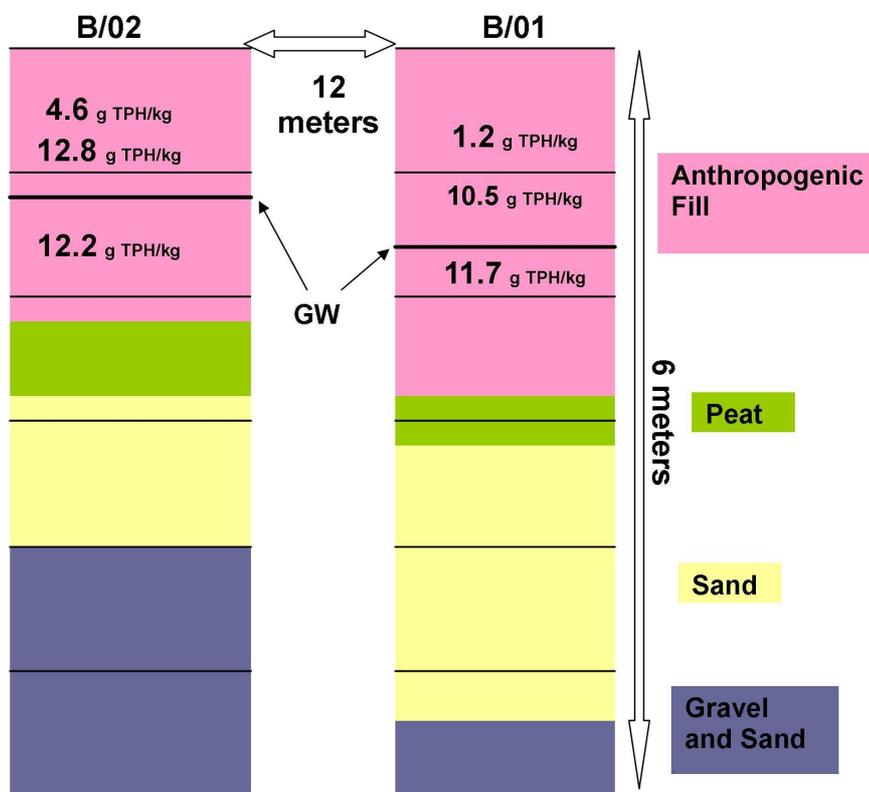


Figure 3. Lithology within source zone at the location of piezometers B/01 and B/02.

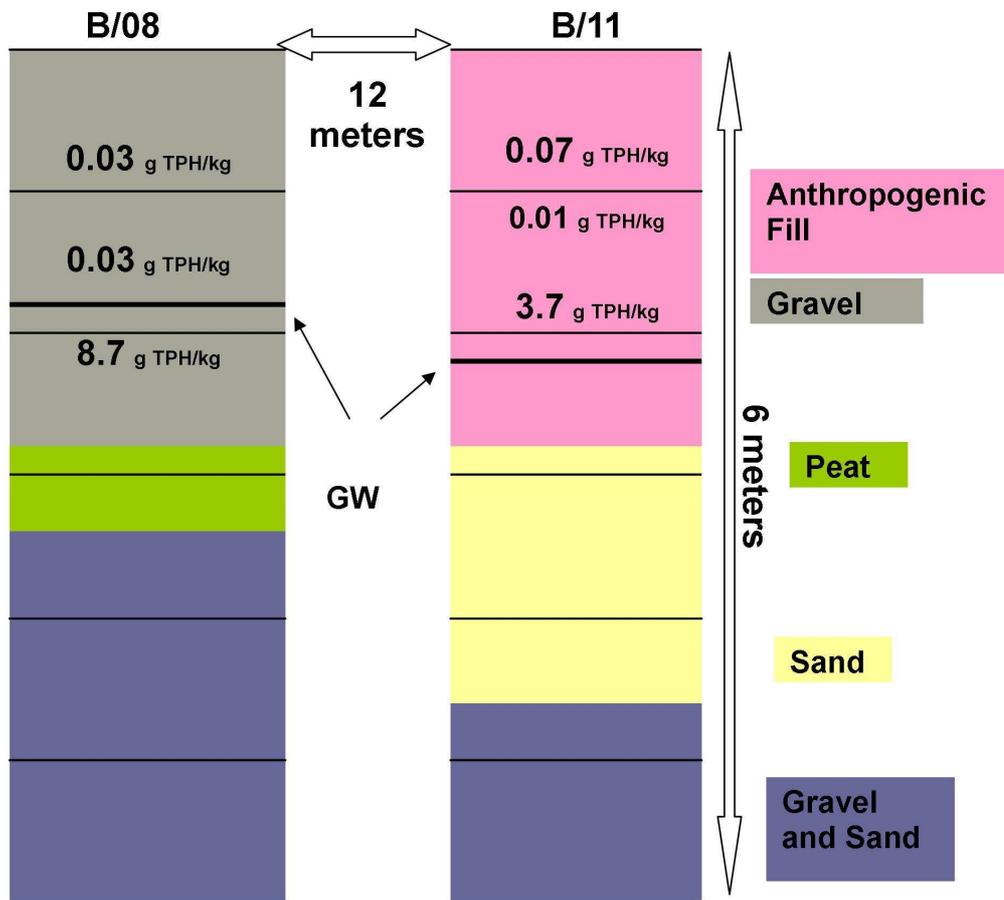


Figure 4. Lithology within plume zone at the location of piezometers B/08 and B/11.

## Results

### *Choice of Sampling Locations:*

For laboratory experiments in which coupled chemical and biological remediation will be tested, samples were collected at the Węglińiec, Poland site included in the EU consortium Upsoil. In order to assess the impact of chemical oxidation and the feasibility of bioremediation under a variety of situations, samples were required from both the source and plume areas. ISCO experiments on source area samples with pure product gives information on the quantity and type of chemical oxidants that effectively reduce contaminant concentrations to levels reasonable for biological polishing of residual pollutants. From plume area samples on which ISCO and/or biostimulation are performed, conclusions can be drawn about the impact of chemical oxidation on the endogenous biodegrading microbial population as well as the natural attenuating ability of this bacterial community. Thus, based on the experimental goals, material from the source and plume is required.

As background information indicated high heterogeneity, especially in the upper anthropogenic layer, multiple samples were required from the source and plume. Through digging at a variety of locations, it can be ensured that results are not related to an anomaly in the material at that specific location. Additionally, determination of remediation feasibility in a variety of soil matrixes necessitated samples from the various lithological layers. Consumption of ISCO chemicals, the abundance of nutrients to bacteria, sorption and thus availability of the

contaminant, and soil permeability is highly dependent on the type of soil material. In the Węglińiec case, previous work indicates that heterogeneous anthropogenic fill, peat, clay, and sand layers are present in both source and plumes areas of the site (Figures 3 and 4).

Finally, control samples of uncontaminated material for each soil type were also required. Control material gives an idea of the natural situation at the site prior to contamination. Analysis of uncontaminated samples provides both background information on soil characteristics and functionality at the site as well as a broad remediation goal. Control experiments illustrate the impact of chemical oxidation and biostimulation on natural soil matrixes and microbial functionality.

In order to reduce the impact of heterogeneity on results, batch experiments were planned. Although column experiments with undisturbed samples may better represent field conditions where chemical oxidants are injected at one location, results from such experiments are often inconclusive and are difficult to transfer to other soil compositions at other locations. Thus, disturbed samples of each soil type could be collected at each sampling location and homogenized.

#### *Description of Sampling Locations:*

During a sampling trip to Węglińiec, Poland, material from a variety of locations and depths was collected (Table 1). Control, thus uncontaminated material, was collected at various locations within and outside of the terrain. A control sample (W1) was collected to the south just outside of the railway site. This area was not filled with anthropogenic material. Thus the upper layer sampled, starting below the topsoil at a depth of 30 cm, is the same sort of peat as that found deeper at the rest of the site. The location was muddy with tall grass and was adjacent to standing water, which may be the start of the brook. Here both peat above and below the groundwater level (40 cm) and the sand (70-180 cm) were collected. Location W7 was to the east in the original "dune" area of the site where no contamination was observed. Here the upper layer (0-50 cm) of more organic rich brown sand is followed by very light sand down to at least 240 cm (the groundwater level). At location W9 the upper 40 cm were collected as a control sample of the anthropogenic zone. Although this area was far to the east of the source zone and not in the path of groundwater flow, as digging did not continue below 40 cm, it can not be conclusively said that no contamination could occur at this location.

In the source zone of the original contamination, two locations within 15 meters of each other were sampled (W2 and W3, between B/02 and B/01). At both of these locations anthropogenic brown sand was first encountered. At depths between 170 and 230 cm extensive pure product was observed. At W3, a clay layer extending from 80 to 150 cm was found between the top fill and the pure product. Samples were collected from the anthropogenic unsaturated fill layer, the clay (W3 only), pure product, and pure product within the soil matrix.

In the course of sampling, locations outside of the original refueling area were found with pure product. At W6, east of B/04, pure product was encountered at 120 cm following the anthropogenic fill layer. Both the fill and the pure product were sampled. Locations W4 and W5, which were within 5 meters of each other, contained pure product at around 200 cm depth. At these sites, organic rich sand down to 100 or 130 cm is followed by 30-80 cm of clay or peat. In the case of W5, further digging revealed a peat layer (200-230 cm) and sand layer (starting at 230 cm) below the pure product. At these two locations separate samples were collected from each layer and were not combined between holes for a total of 8 containers.

Two locations were sampled for the plume area. At W8, at the north side of the site, contamination was noted at 160 cm and a sample from these 20 cm was collected. Throughout the profile, various layers of sand, gravel, and stones were found and sampled. The groundwater level was not reached. At location W10, contamination was observed at 190 cm, which coincided with the groundwater level. Above the groundwater level, samples of the brown sand layer (80-190 cm below the surface) were collected.

Liquid samples were collected at a variety of sites. Pure product samples were collected in glass bottles from the following piezometers: B/03, B/10, B/02, S10, B/06, S9, and S11. Additionally, at W3 and W6 pure product sample was collected. The chromatographs of these samples, as analyzed by GC-FID, will be used to determine the extent of weathering of the diesel pure product. The brook exits the north side of the site, acting as both a drain and receptor of

Table 1. Location and lithology of sample locations at Węglińiec, Poland.

Location Name	Location Description	Groundwater level	Depth: Sediment Description
W1	Control: No anthropogenic fill	40 cm	0-30 cm: organic topsoil with roots
			30-70 cm: peat
			70-180 cm: (coarse) sand, brown, some sections become red upon oxidation in air
W2	Source	230 cm	0-100 cm: dark organic rich sand
			100-200 cm: sand
W3	Source	170 cm	0-80 cm: anthropogenic fill with high organic concentration
			80 -150 cm: clay layer
			150-170 cm: pure product
			170-260 cm: contaminated zone with high natural organic matter concentration
			260-280 cm: peat
W4	Plume	200 cm	280-? cm: sandy peat
			0-100 cm: anthropogenic fill
			100-180 cm: peat/organic rich
W5	Plume	200 cm	200 -? cm: pure product in organic rich matrix
			0-130 cm: organic rich
			130-160 cm: clay
			160-200 cm: pure product
W6	Plume	120 cm	200-230 cm: organic peat
			40-120 cm: anthropogenic fill
			120 cm: pure product found
W7	Control: Dune area	240 cm	0-50 cm: organic brown sand
			50-240 cm: light sand, nearly organic free
W8	Plume	not found down to 180 cm	0-40 cm: stones/gravel
			50-120 cm: sand
			120-160 cm: gravel
			160-180 cm: contaminated gravel
W9	Control: Anthropogenic Fill	not found	0-10 cm: gravel/stones
			10-40 cm: sandy anthropogenic fill
W10	Plume	190 cm	0-80 cm: stony anthropogenic fill
			80-190 cm: dark brown sand
			190-? cm: contaminated and organic rich
W11 and W12	Receptor: Brook		

contamination. Here, water was collected at two locations, W11 and W12. Comparison of the GC chromatographs of these samples with that of the soil of the site should give information on the movement of the overall contamination as well as the mobility of specific components. Finally, at each location samples were snap frozen in the field for molecular analysis of the microbial population.

## Conclusions

The proper choice of samples for any laboratory study determines the success and relevancy of the subsequent results. In selecting material, the type of experiments, setup thereof, and research goals must be taken into account. In the case of the Upsoil location Węgliniec, Poland, samples were collected for experiments in which the feasibility of coupled chemical oxidation and bioremediation will be investigated.

Previous monitoring and investigations gave background information on the history and extent of contamination at the site. Based on information gleaned from previous contaminant and sediment characterizations, sampling sites were chosen. Material was collected from the source and plume areas, as well as from uncontaminated control locations. From each sampling location, material was taken at a variety of depths to ensure that experiments could be performed on a variety of soil matrixes. Integration of results from this site characterization with future experiments on bioremediation in chemically pretreated soils will provide information on the optimization of (coupled) *in situ* remediation technologies and their application in this field.

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