Field-scale Assessment of Phytoremediation of a Flare-Pit Soil in Carlyle, SK

Year 4 Overview

Prepared by:

Richard E. Farrell
SAF Research Chair–Soil Biological Processes
Department of Soil Science
University of Saskatchewan
51 Campus Drive
Saskatoon, SK Canada
S7N 5A8

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Talisman Energy
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This document presents a summary of data obtained during the phytoremediation trials undertaken at the Talisman Energy site located in Carlyle, SK. This work was conducted over four field seasons during the period from May 2002 through October 2005. Research at the Talisman site was conducted under the umbrella of a much larger (NSERC funded) project to assess the effectiveness of phytotechnologies for reducing contaminant levels to environmentally acceptable endpoints (as defined by the Canadian Council of Ministers of the Environment; CCME). As such, this work will eventually be placed in context with other (on-going) field and laboratory studies once they are completed. A final report on the phytotechnology research will be issued in mid-2007.

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Major support for this project was provided through a NSERC–Strategic Program Grant (Germida, Farrell et al. 2003–2006) with additional funding obtained through an initiative facilitated by PTAC Petroleum Technology Alliance Canada. Funding agencies included Environment Canada; the Canadian Association of Petroleum Producers (CAPP) Environmental Research Advisory Council; the Program of Energy Research and Development (PERD); Talisman Energy; and the Saskatchewan Agriculture & Food Strategic Research Program in Soil Biological Processes. In-kind support was generously provided by Talisman Energy and their environmental contractor, Strata Environmental Ltd.

Executive Summary

A cooperative trial to test the use of vegetation to enhance treatment of soils contaminated with weathered petroleum hydrocarbons (PHCs) was initiated in 2002. Experimental protocols were adapted from those developed by the *Phytoremediation Action Team* of the *USEPA Remediation Technologies Development Forum*. Four treatments were compared in a randomized complete block experimental design with four replications. Treatments included (1) an unplanted, unfertilized control; (2) an unplanted, fertilized control; (3) a standard cool-season grass/legume mixture composed of a combination of fescue, ryegrass, and a legume; and (4) a locally optimized treatment that included slender wheatgrass, tall wheatgrass, Altai wild rye, red clover, and Nuttall's salt-meadow grass. Each trial was monitored for a three growing seasons, with soil sampling conducted at planting and at the end of each growing season. Soils were sampled at two depths (0–15 cm and 15–45 cm). Soil samples analyzed for total petroleum hydrocarbons (TPH), polynuclear aromatic hydrocarbons (PAHs), and CCME PHC-fractions using standard (CCME) methods; plant assessments (above- and below-ground biomass) were conducted at the end of each growing season.

Sampling at the time the site was established (t_0) yielded a mean TPH concentration of 5551 mg kg^{-1} (0–45 cm). At the end of the fourth growing season (October 2005) TPH concentrations averaged across the site had been reduced to 1101 mg kg^{-1} in the surface (0–15 cm) soil and 1942 mg kg^{-1} in the sub-surface (15–45 cm) soil. Reductions in PHC concentrations in the planted plots and unplanted plots were generally similar, though reductions in the F3 (C16–C34) fraction generally occurred more rapidly in the planted treatments. Likewise, reductions in PHC concentrations were generally greater in the plots amended with fertilizer and compost. There were no significant differences between the site-specific (USASK) and standard (RTDF) plant mixes; likewise, there were no significant differences between the planted and unplanted treatments. [Note: differences between treatment means were assessed using the least significant difference test at a probability level of $P \leq 0.20$.]

What is the purpose of the project?

Phytotechnologies involve the plant–assisted bioremediation of organic and inorganic contaminants and are essentially a form of ecological engineering that depends on natural, synergistic relationships among plants, microorganisms and the environment. Since 1998, our research has focused on assessing the effectiveness of phytotechnologies as a means of reducing petroleum hydrocarbon (BTEX, TPH & PAH) concentrations in soils contaminated with weathered oil product. This focus reflects the fact that (i) many types of PHCs are amenable to microbial degradation; (ii) the phytoremediation of organic contaminants often involves enhanced microbial degradation in the rhizosphere; and (iii) there are an estimated 200,000 PHC-contaminated sites in the Prairie provinces alone. During the past several years, the Environmental Biotechnology Applications Division (EBAD) of Environment Canada has been working in partnership with other federal departments, provincial governments, universities (including the University of Saskatchewan) and the private sector to assess the utility of plants in a remediation capacity under prevailing Canadian environmental conditions and associated regulatory oversight. The results of this work indicate that to fully exploit and use phytoremediation we need to gain a better understanding of: (i) the pool of phytoremediation species found in Canada; (ii) how phytoremediation operates under unique Canadian climatic conditions; (iii) the mechanisms employed by phytoremediator plants to restore contaminated sites; and (iv) the agronomic requirements needed to maximize phytoremediation as an efficient and cost-effective cleanup technology. So, while there is clear recognition that phytotechnologies have the potential to play an important role in future remediation strategies in Canada, there remains a ***critical need*** for 'field performance data' to verify this potential, as well as to assess its limitations and determine appropriate uses of the newly emerging phytotechnologies.

To address this need, we have been working with our industry and government collaborators to conduct field-scale assessments of phytotechnologies at sites impacted with weathered hydrocarbons in the oil and gas producing regions of Saskatchewan and Alberta. The long-term objective of this study is to assess and demonstrate the utility of phytoremediation as a means of reducing petroleum hydrocarbon levels in oil-contaminated soils to environmentally acceptable endpoints.

Research Plan and Methodology

Field sites were established in Carlyle, SK (Talisman Energy) in the spring of 2002 and in Bruderheim, AB in the spring of 2003. The Carlyle site consisted of oil-impacted soil from a buried flare pit, whereas the Bruderheim site consisted of PHC-contaminated soil from a tank battery (formerly located at the same site). Both sites were selected following conversations involving members of the *U of S* phytoremediation research team, the PTAC—Phytoremediation Steering Committee, Environment Canada, and our industry collaborators.

This report deals exclusively with research conducted at the Carlyle (Talisman Energy) site. General climate and site conditions are summarized in Table 1. Activities, milestones and timelines are summarized in Table 2.

Protocols used to evaluate the effectiveness of the plant-based remediation strategy (enhanced rhizosphere biodegradation) implemented at this site were adapted from those

described in the *Phytoremediation of Petroleum Hydrocarbons in Soil Field Study Protocol* developed by the USEPA Remediation Technologies Development Forum (RTDF)–*Phytoremediation of Organics Action Team*. The complete protocol can be viewed at the RTDF website (**http://www.rtdf.org/public/phyto/protocol/protocol99.htm**).

Table 1. Summary of climate and site conditions at the Carlyle phytoremediation site.

	Carlyle (Site L)*
Ecozone	Mixed grassland/Parkland
Soil type	Dark Brown/Black Chernozem
Soil texture	Clay
Analytical: TPH (mg kg^{-1}): 0–45 cm	5510
Mean annual precipitation (mm)	422
Growing season length (days)	125
Average last frost	May 19th
Average first frost	September 21st
Depth to groundwater (m)	5–6
Contaminant source	Flare-pit soil
Depth of contamination (cm)	45

* RTDF designation.
§ Based on results of the initial (t_i) grid sampling.
¥ Depth of contaminated soil in the raised bed.

Table 2. Sampling schedule (activities, milestones, and timelines).

Sampling Event	**Timing & Purpose**	**Analysis**	**Date completed**
t_0	Characterize site variability	TPH, PHC-fractions, PAHs; total metals; salinity	03 Nov 2001
$t_{1.1}$	After tillage but before planting: – determine fertilizer requirements	Fertility testing*, TPH & PHC-fractions	23 May 2002
$t_{1.2}$	End of 1st growing season: – assess PHC degradation	TPH & PHC-fractions; plant assessments	22 Oct 2002
$t_{2.1}$	Start of 2nd growing season: – assess PHC degradation	Fertility testing	05 May 2003
$t_{2.2}$	End of 2nd growing season: assess PHC degradation	TPH & PHC-fractions; plant assessments	22 Oct 2003
$t_{3.1}$	Start of 3rd growing season: – determine fertilizer requirements	Fertility testing	25 May 2004
$t_{3.2}$	End of 3rd growing season – assess PHC degradation & plant uptake	TPH & PHC-fractions; plant assessments	06 Oct 2004
$T_{4.1}$	Start of 4th growing season: – determine fertilizer requirements	Fertility testing	06 Jun 2005
$T_{4.2}$	End of 4th growing season – assess PHC degradation & plant uptake	TPH & PHC-fractions, PAHs; plant & microbial assessments§	17 Oct 2005

* Includes available N, P, & K; micronutrients, soil organic carbon, and pH; electrical conductivity (EC) and sodium adsorption ratio (SAR) were determined at $t_{1.1}$ and $t_{3.2}$; particle size analysis was performed at $t_{1.1}$ only.
§ Enumeration (using MPN) of TPH degraders.

Initial Site Characterization. Site selection was based, in part, on the criteria outlined in the *Phytoremediation Decision Tree* produced by the Interstate Technology Regulatory Cooperation workgroup [ITRC, 2000]. Initial site characterizations involved collecting and organizing existing information regarding the physical, biological and chemical status of the site, including: location, type of site, source of contamination, age of site, age of contaminant, estimated area, depth and/or volume of contaminated soil; site history (including any treatment with pesticides/herbicides); soil characteristics (soil type & texture; pH & EC; CEC & OM; nutrient, water, and aeration status); and plants present on-site prior to establishment of the research plots.

The research site (located at 5-18-8-3-W2) included sufficient area for the construction of a raised bed system (35-m × 40-m × 0.45-m) to contain the contaminated soil. Source material for the site consisted of *ca*. 300 m^3 of soil contaminated with heavier end PHCs (15,000 mg TEH kg^{-1}), *ca*. 200 m^3 of soil with lighter-end PHCs (233 mg TEH kg^{-1}), and *ca*. 130 m^3 of overburden material (264 mg TEH kg^{-1}). Analyticals for the source materials are summarized in Table 3. The site was surrounded by cropland, and was under production during the four years (2001–2005) that this research was conducted.

Table 3. Summary of the analytical data for the flare pit material excavated from the Carlyle phytoremediation research site at 5-18-8-3-W2.

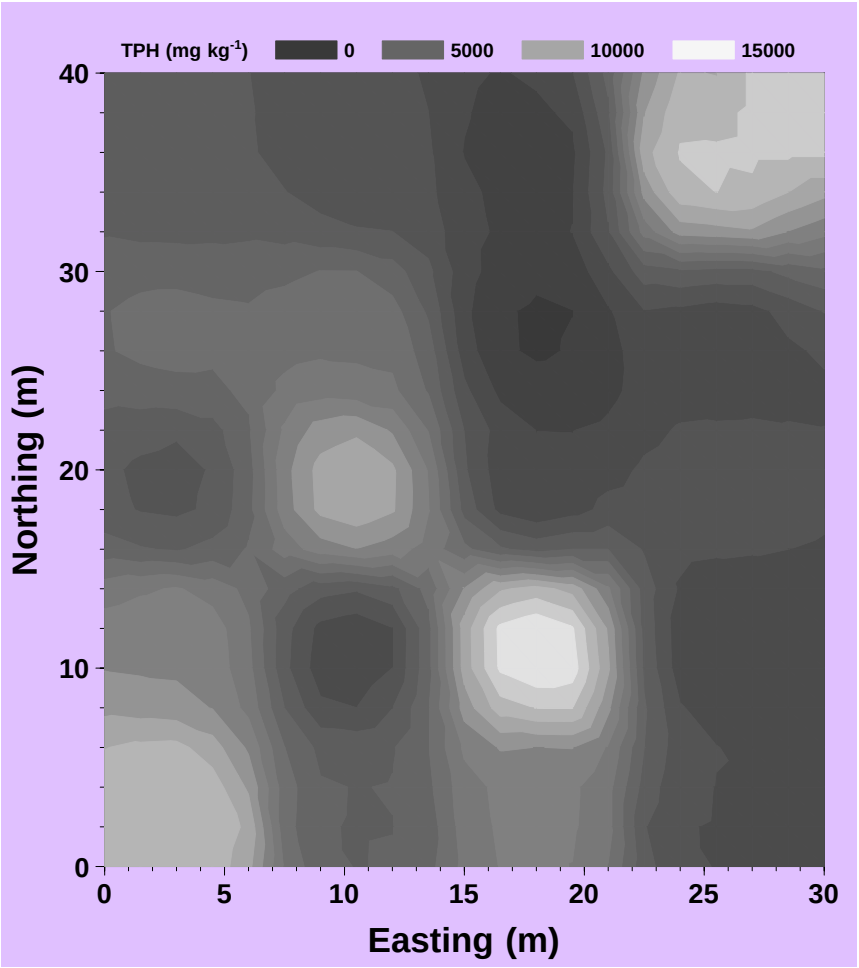
Source Material	Volume (m^3)	EC* ($dS\ m^{-1}$)	BTEX§ ($mg\ kg^{-1}$)		TEH¥ ($mg\ kg^{-1}$)
Pit Material #1	1300	8.2	B	0.58	233
			T	0.60	
			E	2.08	
			X	3.31	
Pit Material # 2	200	59.2	B	21.4	15,000
			T	27.0	
			E	18.7	
			X	32.8	
Overburden	200	17.7	B	<0.01	264
			T	<0.01	
			E	<0.01	
			X	0.09	

* Electrical conductivity. (Note: all analyses were conducted by Enviro-Test Laboratories, Saskatoon, SK.)
§ Benzene, toluene, ethylbenzene, and xylene.
¥ Total extractable hydrocarbons.

Construction of the raised bed system occurred on 30 October 2001. Surface soil from the plot area was removed and used to construct a berm (*ca*. 0.5-m high) around the site; source materials from the buried flare put were then mixed and moved into place using a bulldozer. A rectangular grid (35-m × 40-m; with a 7.5-m grid spacing) composed of 20 sampling sites was established on 03 November 2001; soil samples (0–15 cm and 15–45 cm) were collected using a split-core sampler with a slide hammer and stored on ice for their return to the U of S,

where they were frozen until analyzed. Despite efforts to "homogenize" the blended (contaminated) soil, there remained a large degree of spatial variability (see Fig. 1). Similar spatial patterns were observed for the various CCME PHC-fractions (data not shown).

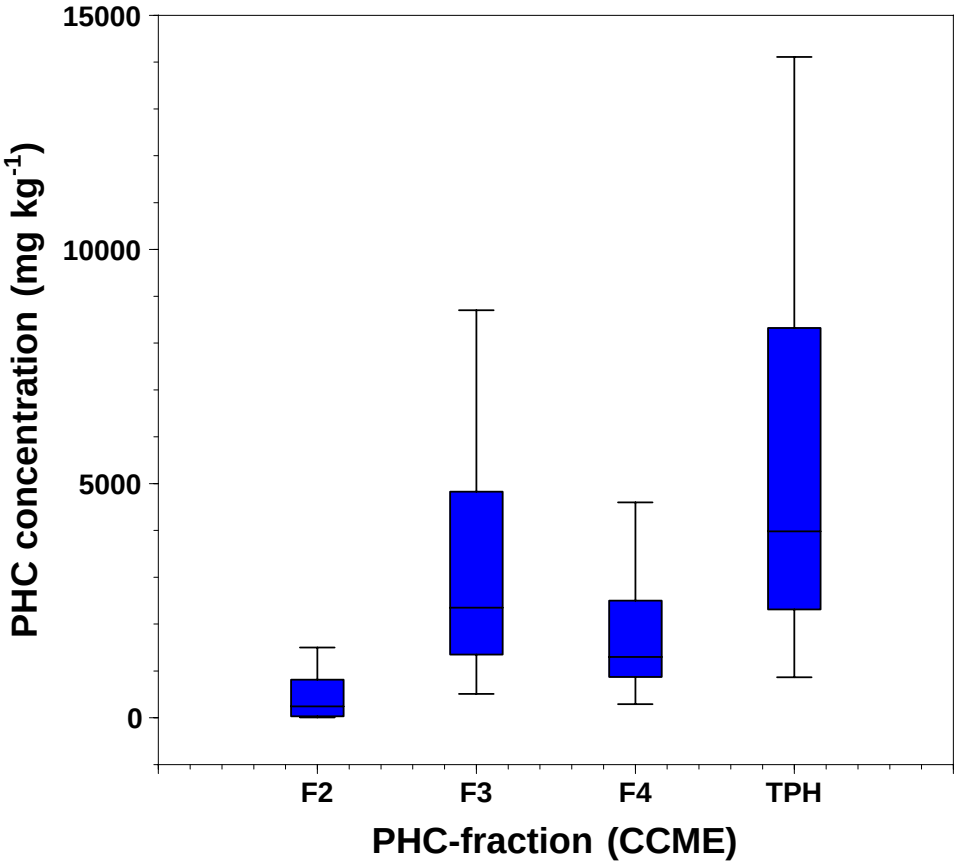
Figure 1. Contour map of total petroleum hydrocarbon (TPH) concentrations in the plot area at the Talisman phytoremediation site (Carlyle, SK).



Whereas all four PHC-fractions exhibited a wide range in concentration (see Fig. 2), only the F3 (C16–C34) and portions of the F1-BTEX fractions (i.e., benzene, toluene, and ethylbenzene) yielded mean concentrations that exceeded the Canadian Soil Quality Guideline (CCME, 2001; CCME, 2004) for a fine-grained soil being remediated for agricultural use.

Due to inclement weather, the addition of soil amendments and establishment of the experimental plots was postponed until spring 2002. During the winter months, however, soil from the Carlyle site was used in growth chamber studies to evaluate soil amendments and conduct plant selection studies. Twenty-one plant species were evaluated for their ability to germinate and grow in the contaminated soil, with six species subsequently being selected for use in the field study.

Figure 2. Distribution of petroleum hydrocarbons (PHC) in the plot area at the Talisman phytoremediation site (Carlyle, SK). The PHC fractions are based on CCME (Canadian Council of Ministers of the Environment) guidelines; F1 = C6–C10, F2 = C10–C16, F3 = C16–C34, F4 = C34–C50.



Soil amendments (gypsum, straw, compost & fertilizer) were required to establish a suitable plant growth environment, and were intended to (i) alleviate salt effects (EC = 5.9 ± 4.8 dS m^{-1}; SAR = 25 ± 12), (ii) improve soil structure and water holding capacity (the structure of the soil was 'massive', resulting in extremely low infiltration rates and water storage), (iii) increase the organic matter content of the soil (the blended flare pit 'soil' consisted primarily of B and C horizon material, with very little native organic matter), and (iv) improve the fertility status of the soil (esp. in terms of N & P fertility). Based on the results of the plant screening, and the requirements of the RTDF protocol, six plant species were selected for use in the field: perennial ryegrass, altai wildrye, creeping red fescue (var. Laurel), tall wheatgrass, Nuttall's salt-meadow grass, and yellow sweet clover[1].

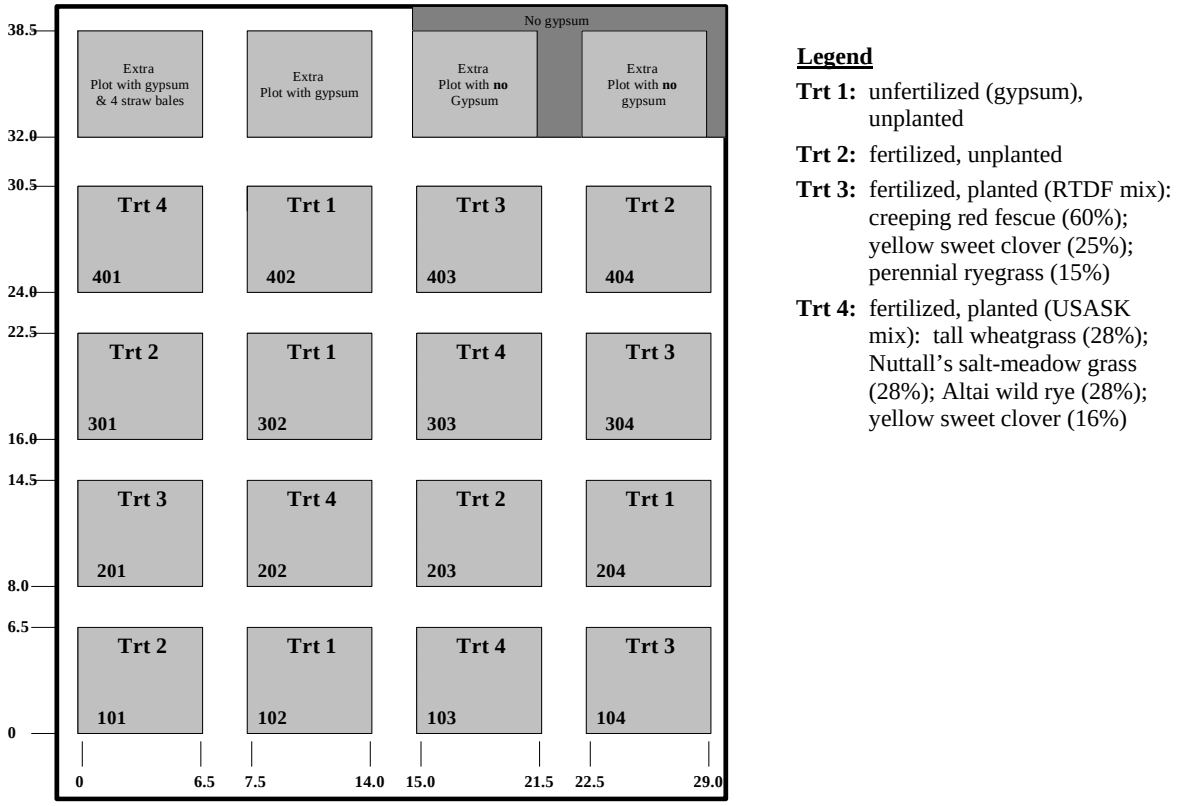
The PHC-contaminated soil (i.e., the blended pit material) was a saline-sodic, heavy clay loam that contained little organic-C (average C:N ratio = 53:1) and was severely deficient of plant available N and P. Moreover, compaction during the construction phase of the raised

[1] Note: alfalfa (var. Rambler) was originally chosen as the legume of choice; however, a labelling error by the seed distributor resulted in our receiving yellow sweet clover. This error was not noted until mid-season, when the plants began to flower.

bed resulted in a soil with massive structure and low infiltration rates. To address salinity issues, the entire plot area received an application of gypsum (50 Mg ha^{-1}); fertility issues were addressed by applying the required amendments to the individual experimental plots.

Experimental Field Design. The experimental design for the Carlyle site was a randomized complete block (RCBD) with four treatments replicated four times (see Fig. 3).

Figure 3. Plot plan for the Talisman phytoremediation site (Carlyle, SK).



Treatments. The experimental treatments included one set (n = 4) of unfertilized/unplanted control plots, one set of fertilized/unplanted control plots, and two sets of fertilized/planted treatment plots. One set of the treatment plots was seeded with a standardized plant mix (identified as the RTDF mix, which was planted at all 13 RTDF sites). The second set of treatment plots was seeded with a site-specific mix of local native/adapted species selected from the plant screening program at the University of Saskatchewan. Individual research plots were *ca*. 6.5-m × 6.5-m.

Soil structure in the amended plots (Trt 2–4) was modified by the incorporation of straw (16 Mg ha^{-1}) and manure (225 Mg ha^{-1}; *ca*. 5-yr old). Fertility issues were addressed by the application of urea (34-0-0; 264 kg ha^{-1}) and $(NH_4)H_2PO_4$ (12-51-0; 96 kg ha^{-1}) fertilizers. Soil amendments were applied and worked into the contaminated soil between May 22nd and

June 5th 2002. The resulting amended soil had a C:N ratio of *ca*. 17:1.

Plots receiving the standard (RTDF) plant mix were seeded with creeping red fescue (47 kg ha^{-1}), yellow sweet clover (19 kg ha^{-1}), and perennial ryegrass (12 kg ha^{-1}). Plots receiving the localized (USASK) plant mix were seeded with tall wheatgrass (81 kg ha^{-1}), Nuttall's salt meadow grass (81 kg ha^{-1}), Altai wild rye (81 kg ha^{-1}), and yellow sweet clover (46 kg ha^{-1}). Because germination rates are generally lower in saline and PHC-contaminated soil than in uncontaminated soil, all plant species were seeded at 4-times the recommended rate. At seeding (05 June 2002), the amended plots (Trt 2–4) received an additional topdressing of composted manure (396 Mg ha^{-1}; *ca*. 20-yr old) into which the seeds were sown. The plots were then packed using a drum roller.

Plot Maintenance. Routine plot maintenance involved the application of fertilizer (based on the results of soil fertility tests conducted at the start of each season); reseeding bare patches in the planted treatments (Trt. 3 & Trt. 4); weed control operations, which involved tillage and spraying the alleyways and berms and hand-weeding the treatment plots; and soil and plant sampling in the fall.

In YEARS 2 and 3 (2003 and 2004) the planted treatment plots received split applications of Scotts® Turf Builder® (29-3-4-6; 352 kg ha^{-1}), with one-half the fertilizer applied at the start of the growing season and one-half applied at mid-season. The plots also received an additional application of a slow release (32-4-8; 320 kg ha^{-1}) fertilizer at the end of each growing season. In YEAR 4 (2005), the plots received a single application of Scotts® Turf Builder® (125 kg ha^{-1}) at the start of the season.

Based on visual observations of percent ground cover and species composition, the planted plots (Trt. 3 & 4) were reseeded as necessary to fill in bare areas, while keeping the initial (desired) composition of the plots the same. Bare areas in the plots were reseeded following the 2002 (YEAR 1) and 2003 (YEAR 2) growing seasons. Plots received one-half the required amount of seed in the fall (following soil and plant sampling) and the remaining one-half the following spring (at the same time the first split-application of fertilizer was applied).

Site Sampling and Analysis. The initial soil characterization and yearly fertility assessments were carried using standard methods [Carter, 1993]. Plant assessments (percent cover, shoot height, rooting depth & density) were carried out at the end of each growing season, with hydrocarbon analysis of plant tissues being conducted in the last year (i.e., at $t_{3.2}$) only. Soil and plant samples were analyzed for PHCs using accelerated solvent extraction (*modified EPA Method 3541*) followed by analysis for the following target classes: (i) total petroleum hydrocarbons (TPH) by GC-FID (*EPA Method 8015*); PAHs by GC-MS (*EPA Method 8270*)[2]; (ii) CCME PHC-fractions by GC-FID (fractions F1–F4) and GC-MS (fraction F1-BTEX) (*Reference Method for the Canada-Wide Standard for Petroleum Hydrocarbons in Soil - Tier 1 Method;* CCME, 2001); and (iii) biomarker steranes and triterpanes (e.g., hopane

[2] The initial (t_0) site characterization revealed that PAHs were present at very low concentrations, if at all. Therefore, due to cost constraints, PAH analyses were repeated only in YEAR 3 (i.e., at $t_{3.2}$). Likewise, PHC concentrations in the sub-surface (15–45 cm) were quantified in only the first and last growing seasons.

or norhopane) by GC-MS (*modified EPA Method 8270*)[3]. Microbial assessments also were carried out at the end of each growing season using the most probable number (MPN) technique to enumerate populations of general hydrocarbon (alkane) degraders, diesel degraders, and PAH degraders.

A thin-walled, split-core sampler tube fitted with a stainless steel liner was used to collect soil samples. Eight random sub-samples from each treatment plot were combined to make one composite sample per plot. Replicate (n = 5) samples of each composite soil were packed[4] into 250-mL wide-mouth, clear-glass jars with Teflon-lined polypropylene lids (e.g., VWR TraceClean™–QA), stored on ice in a cooler, and transported to the University of Saskatchewan where they were placed into a -20°C freezer and stored till they were analyzed using the methods described above.

Plant assessments were conducted at the end of each growing season. These included visual assessments of bare ground and plant cover and the percent of plant cover occupied by each species. In addition, the above-ground biomass production was assessed by harvesting the vegetation covered by three randomly placed 0.5-m × 0.5-m quadrats per plot. Plants were harvested to level of 1-2 inches from the ground surface so as not to harm the crowns. Plant samples were placed in paper bags, dried, and weighed.

Root samples were obtained by collecting one 5-cm (i.d.) soil core from within each of the three quadrats selected for biomass production. A single 0–45 cm core was collected and then split into two sections (0–15 cm and 15–45 cm); each section of the soil cores were then placed into a Ziploc bag, labelled, and placed on ice for transport to the University of Saskatchewan. The cores were processed to separate the roots from the soil for determinations of root biomass, root length density, and average root diameter. The roots were cleaned thoroughly and scanned to obtain a digital image of the roots. Digital images were then processed using *ScanTastic* (ver. 4) software (Second Glance Software, Bremerton, WA) to estimate root length, root diameter and the estimated root length density. Following the digital image analysis, the roots were dried and weighed to obtain an estimate of root biomass.

Climate data (daily precipitation and temperature) were obtained from Environment Canada; daily average temperatures and cumulative growing season precipitation recorded at Carlyle during the study period (March 2002–October 2005) are summarized in Fig. 4.

Results

Plant assessments. Ground cover in the planted treatment plots (Trt. 3 & 4) increased each season, ranging from about 35–60% at the end of the first growing season (2002) to about 95% at the end of the fourth growing season (2005) (Figs. 5 & 6). In each of the first three years (2002–2004), the site-specific (USASK) plant mix out performed the RTDF mix; i.e., plant establishment and percent ground cover were significantly greater in plots seeded with the USASK mix (Trt. 4).

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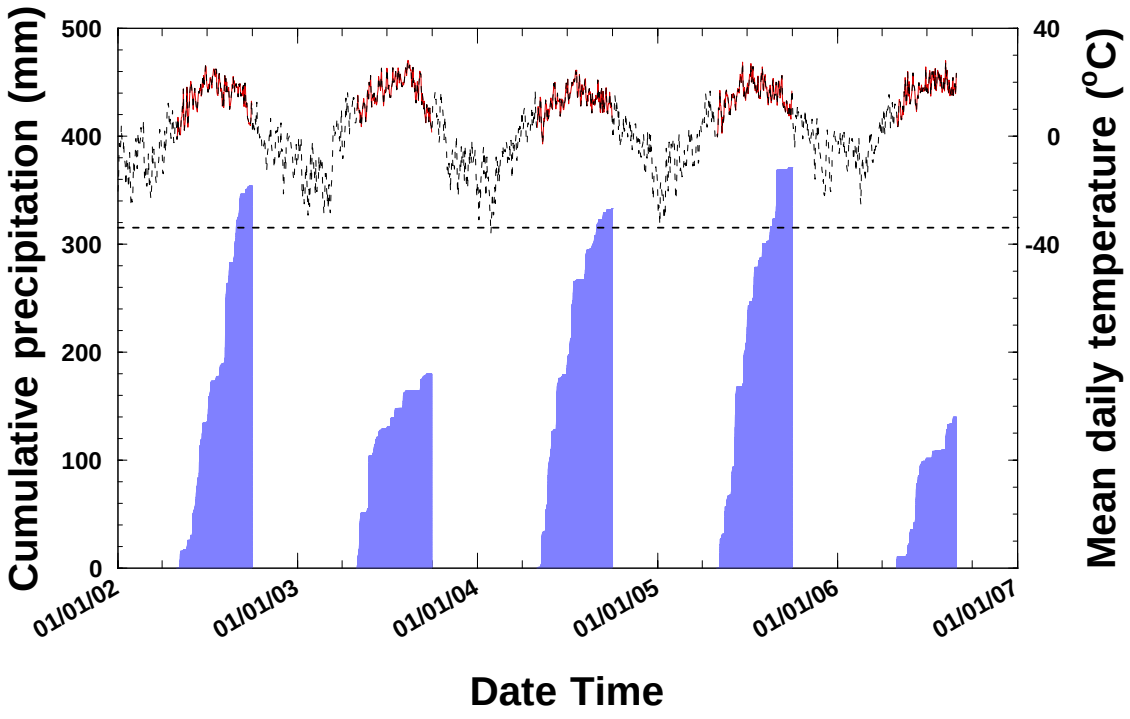
[3] The standard biomarkers were not present in the samples from the Carlyle site; therefore, there has been no attempt to 'normalize' the data against the biomarkers. Though similar difficulties have been found at the other RTDF sites, this work is continuing.

[4] The jars were packed so as to eliminate any headspace.

Figure 4. Climate data for Carlyle, SK for the period from January 2002 through August 2006. (Data obtained from Environment Canada.) Note: cumulative precipitation curves are for growing season precipitation only; average daily temperatures during the growing season are shown as red symbols.



Despite efforts to maintain the initial species composition of the planted treatments, significant shifts in species composition were observed in both the RTDF (Trt. 3) and USASK (Trt. 4) treatments (see Figs. 5 & 6, respectively). In plots seeded with the RTDF mix, creeping red fescue (CRf), which made up 60% of the initial seed mix, had all but disappeared by the end of the second growing season. Yellow sweet clover (Ysc) was the most successful species during the first three growing seasons (accounting for 45–60% of the groundcover, but decreased in importance as the perennial ryegrass (PR) became better established. Indeed, by the end of the fourth growing season (2005), perennial rye was the dominant species in the RTDF plots–accounting for more than 80% of the vegetative cover.

Significant changes in species composition also were observed in plots seeded with the USASK mix (Trt. 4; Fig. 6). In these plots, tall wheatgrass (Twg) had become the dominant species during the second growing season and, by the end of the fourth growing season, accounted for about 96% of the vegetative cover in the plots. Conversely, the Nuttall's salt-meadow grass (Nsmg), Altai wild rye (Awr) and yellow sweet clover (Ysc) combined comprised less than 5% of the species present in the plots. In general, the observed shifts in species composition in both the RTDF and USASK plots reflect inherent differences in competitiveness among the plant species and the ability of the species to adapt to changing environmental conditions.

In terms of biomass production, the site-specific (USASK) mix generally out-performed the RTDF mix (Figs. 7 & 8); this was especially true during 2004 and 2005. Low biomass

yields during the 2002 and 2003 seasons (< 1000 kg ha^{-1} for the RTDF mix and < 2000 kg ha^{-1} for the USASK mix) reflected poor weather conditions and a significant grasshopper infestation that prevailed during the first two years of the study. The 2004 season was characterized by good weather (warm with adequate moisture) which resulted in significant increases in biomass production–especially in the plots seeded with the site-specific (USASK) mix. Biomass yields during the fourth season (2005) increased in the plots seeded with the RTDF mix (Trt. 3), but decreased in the plots seeded with the USASK mix (Trt. 4). These results reflect the combined effect of weather and the change in species composition. Increased biomass production in the RTDF plots also reflect the significant (25%) increase in stand establishment that occurred during the 2005 season.

Figure 5. Plant assessment summary for plots seeded with the standard RTDF plant mix (Trt. 3). Plant samples were collected in October of each year.

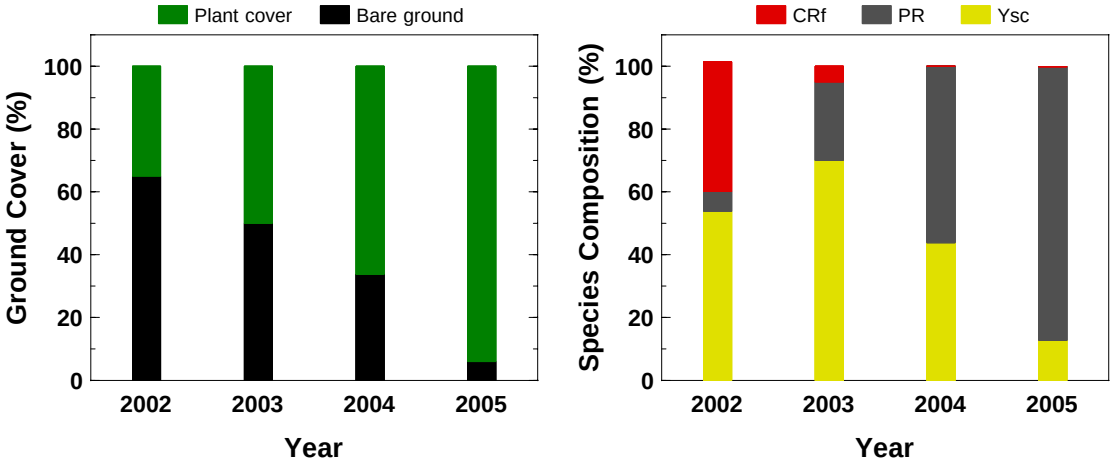
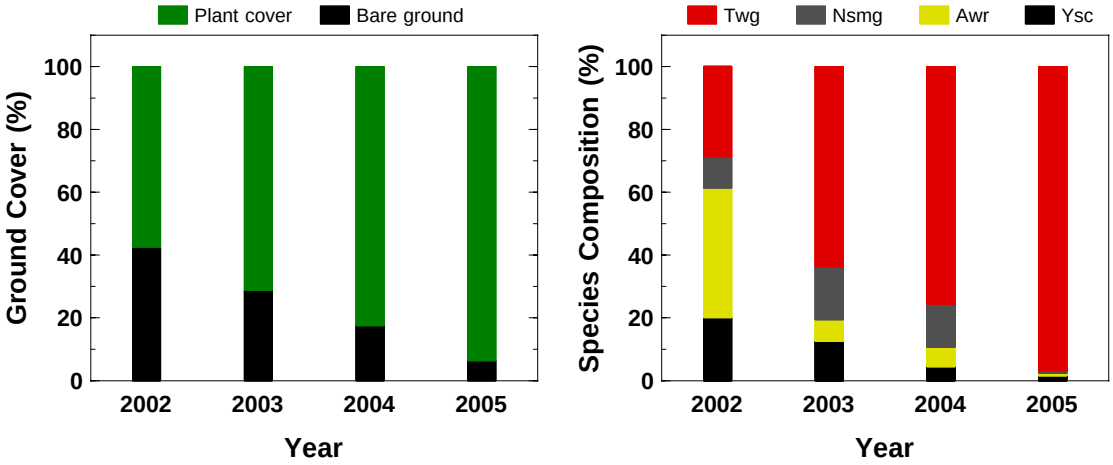


Figure 6. Plant assessment summary for plots seeded with the localized USASK plant mix (Trt. 4). Plant samples were collected in October of each year.



Root production in both planted treatments increased steadily during the first three seasons (Figs. 7 & 8), reflecting increases in both plant cover and overall plant growth. Below-ground biomass production during the 2005 season was only slightly greater than that measured at the end of the 2004 season. Over time, it also was observed that the roots tended to extend deeper into the sub-surface (i.e., into the 15–45 cm depth increment) and that this effect was generally greater in soils from plots seeded with the USASK plant mix (data not shown).

Figure 7. Above- and below-ground biomass production in plots seeded with the standard RTDF plant mix (Trt. 3). Root density values are for the 0–15 cm depth increment. Plant samples were collected in October of each year.

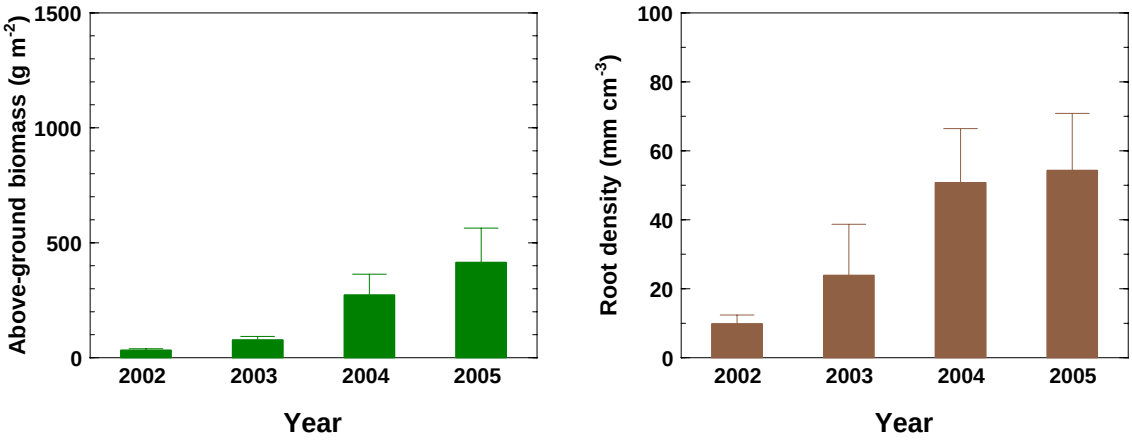
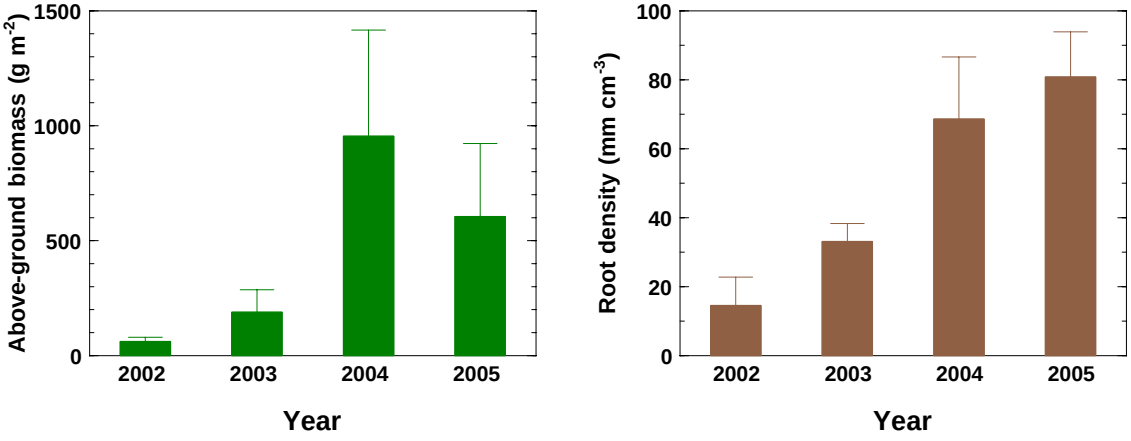


Figure 8. Above- and below-ground biomass production in plots seeded with the localized USASK plant mix (Trt. 4). Root density values are for the 0–15 cm depth increment. Plant samples were collected in October of each year.



Microbial assessments. Populations of indigenous PHC-degraders were present in the contaminated soil, but were generally present at significantly greater numbers in the

straw/manure amended and fertilized soils (see Figs. 9 & 10). In general, population shifts over time for all segments of the degrader population followed the same general pattern regardless of treatment. That is, there was a significant decline in population numbers from the first to the second growing season (2001 to 2003); followed by a small increase and levelling off of the population numbers in the third (2004) and fourth (2005) growing seasons. Whereas these trends were independent of treatment for the diesel- and alkane-degrader populations, the presence of plants seemed to have a beneficial effect on the PAH-degrader populations. That is, in both the surface (0–15 cm) and sub-surface (15–45 cm) soils, PAH-degrader populations in the unplanted treatments (Trt. 1 & 2) decreased during the second growing season and remained relatively constant thereafter. In the planted treatments (Trt. 3 & 4), however, PAH-degrader populations increased significantly during the third and fourth growing seasons–equalling or exceeding the initial population numbers. Moreover, averaged across growing seasons, the PAH-degrader populations in the planted treatments generally exceeded those in the unplanted treatments. This was especially evident in the sub-surface, and is believed to reflect the influence of the roots (and root exudates) on PAH bioavailability.

Figure 9. PHC-degrader populations (MPN data) in soils (0–15 cm) from the Talisman phytoremediation site at Carlyle, SK. Trt. 1 = unamended, unplanted control; Trt. 2 = amended, unplanted control; Trt. 3 = amended and planted with the RTDF plant mix; Trt. 4 = amended and planted with the USASK plant mix.

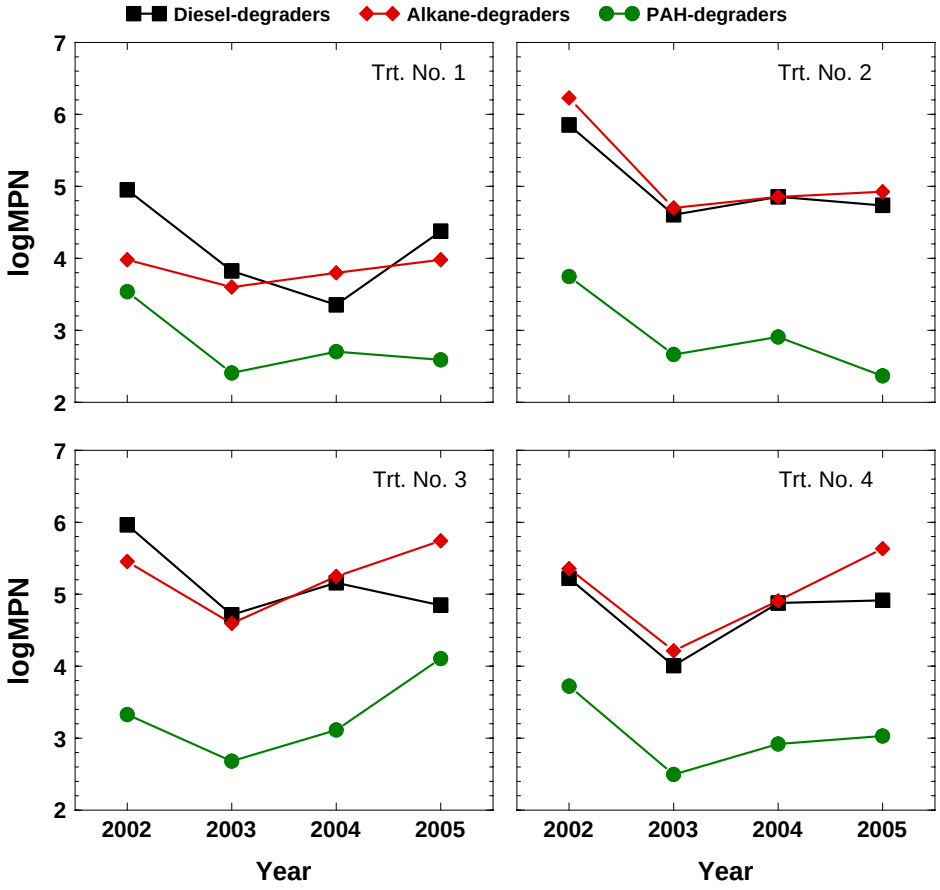
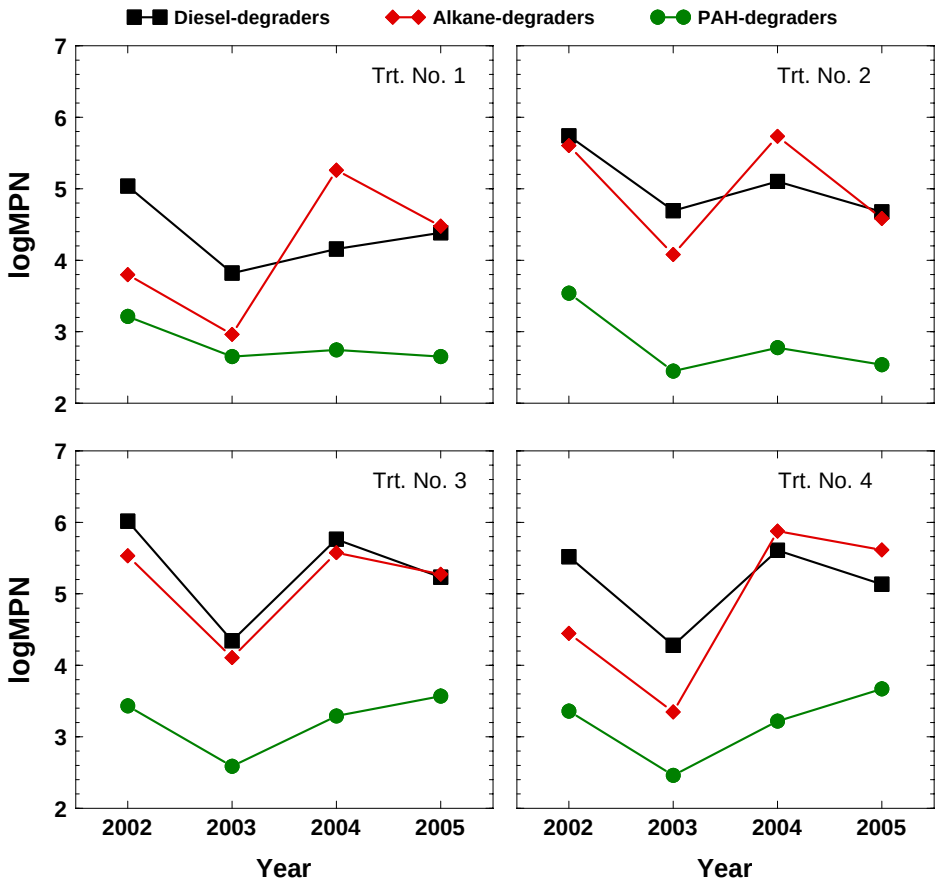


Figure 10. PHC-degrader populations (MPN data) in soils (0–15 cm) from the Talisman phytoremediation site at Carlyle, SK. Trt. 1 = unamended, unplanted control; Trt. 2 = amended, unplanted control; Trt. 3 = amended and planted with the RTDF plant mix; Trt. 4 = amended and planted with the USASK plant mix.



PHC degradation. Initial soil characterization of the Carlyle site revealed that (i) the average TPH concentration in the excavated flare pit soil was 5551 mg kg^{-1} (Note: the soils were sampled after construction of the raised bed was completed, but before amendments were applied to the treatment plots); (ii) benzene, toluene, and ethylbenzene were present at levels in excess of the Canadian soil quality guidelines (SQG) for a fine-grained, agricultural soil; (iii) only the F3 (C16–C34) PHC-fraction exceeded the SQG for a fine-grained agricultural soil; (iv) naphthalene and phenanthrene were the only PAHs detected that exceeded the SQG; (v) trace metal concentrations were within the range commonly found in soils and were generally well below the SQGs; and (vi) the soil was classified as saline-sodic, with an electrical conductivity (EC) and a sodium adsorption ratio (SAR) that exceeded the SQGs. The primary soil chemical indicators used to describe the Carlyle soil are summarized in Table 4.

Upon completion of the fourth growing season (October 2005) TPH concentrations averaged across the site had been reduced to 1101 mg kg^{-1} in the surface (0–15 cm) soil and 1942 mg kg^{-1} in the sub-surface (15–45 cm) soil. The most volatile PHCs [the F1 (C6–C10) and F1-BTEX compounds] were virtually eliminated by the simple act of mixing and working the soil. Petroleum hydrocarbons in the F2 (C10–C16) fraction also decreased

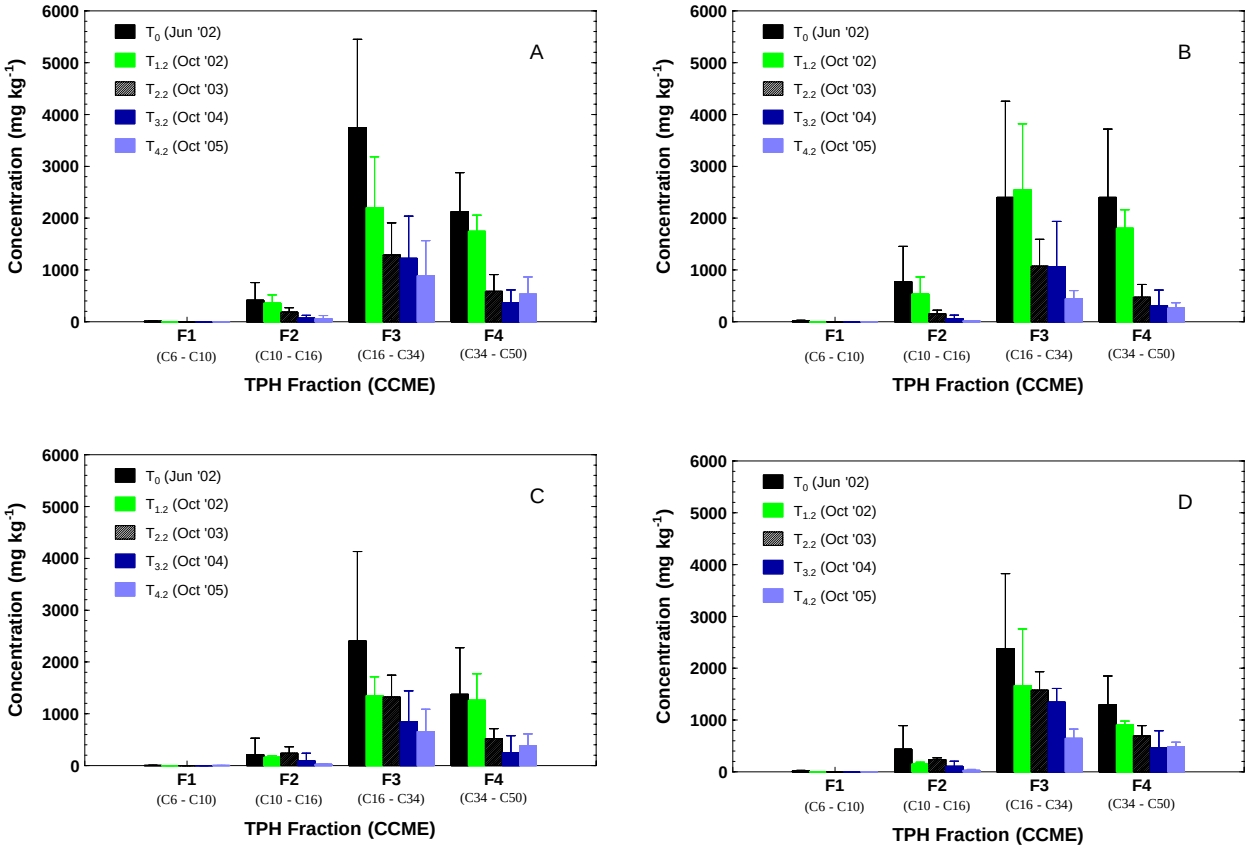
significantly throughout the study (Fig. 11), with less than 10% of the initial F2 fraction remaining after the fourth growing season. Significant reductions in PHC concentration also were observed in the F3 (C16–C34) and F4 (C34–C50) fractions (Fig. 11), with reductions in the F3 fraction resulting in a final, site-averaged concentration that was less than the SQG for the surface 15-cm. However, the site-averaged F3 concentration in the sub-surface (15–45 cm) soil still exceeded the SQG for a fine-textured agricultural soil–though only by about 5%.

Table 4. Primary soil chemical indicators [petroleum hydrocarbons (PHCs), polyaromatic hydrocarbons (PAHs), trace metals, and salinity indices] measured in the surface (0–15 cm) and sub-surface (15–45 cm) soils from the phytoremediation site at Carlyle, SK.

Soil Parameter	SQG* (mg kg^{-1})	- - - Initial (t_0) Conc. - - - (mg kg^{-1}) (0–45 cm)	- - - Final (t_3) Conc. - - - (mg kg^{-1}) (0–15 cm)	(15–45 cm)
PHCs				
F1 (C6–C10)	**260**	8.1 (11.1)	1.0 (0.8)	1.4 (1.9)
F2 (C10–C16)	**900**	454 (526)	31.5 (37.3)	254 (523)
F3 (C16–C34)	**800**	3254 (2294)	657 (463)	834 (947)
F4 (C34–C50)	**5600**	1835 (1224)	411 (248)	853 (886)
BTEX				
Benzene	**0.0068**	0.34 (1.26)	BDL	BDL
Toluene	**0.08**	0.51 (0.99)	BDL	BDL
Ethylbenzene	**0.018**	0.86 (1.34)	BDL	BDL
Xylenes	**11**	4.93 (8.30)	BDL	BDL
PAHs[§]				
Naphthalene	**0.1**	0.23 (0.08)	0.03 (0.01)	0.13(0.01)
Phenanthrene	**0.1**	0.15 (0.06)	0.04 (0.01)	0.04(0.01)
Pyrene	**0.1**	0.08 (0.02)	BDL	BDL
Trace Metals[¥]				
Arsenic (As)	**12**	7.4	ND	ND
Barium (Ba)	**750**	213	ND	ND
Cadmium (Cd)	**1.4**	BDL	ND	ND
Chromium (Cr)	**64**	10	ND	ND
Cobalt (Co)	**40**	26.7	ND	ND
Copper (Cu)	**63**	27	ND	ND
Lead (Pb)	**70**	9	ND	ND
Mercury (Hg)	**6.6**	0.04	ND	ND
Nickel (Ni)	**50**	28	ND	ND
Selenium (Se)	**1**	0.6	ND	ND
Vanadium (V)	**130**	42	ND	ND
Zinc (Zn)	**200**	68.5	ND	ND
pH	**6–8**	7.8 (0.3)	7.6 (0.1)	7.7 (0.1)[⌗]
EC (ds m^{-1})	**2**	5.9 (4.8)	3.9 (3.7)	9.2 (3.6)[⌗]
SAR	**5**	24.9 (12.4)	7.2 (6.5)	15.0 (7.7)[⌗]

***** Canadian Soil Quality Guideline (CCME, 2001; CCME, 2004): Land use = agricultural; Soil texture = fine-grained.
† Below detection limit.
‡ Not determined.
§ No other priority PAHs were present in the soil.
¥ Composite sample for the 0–45 cm depth.
⌗ Samples analysed for the control (Trt. 1) plots only.

Figure 11. Petroleum hydrocarbon (PHC) concentrations (mg kg^{-1}) in the upper 15 cm of soil in the experimental plots at Carlyle, SK. (A) Treatment 1: unplanted, unfertilized; (B) Treatment 2: unplanted, fertilized; (C) Treatment 3: fertilized, planted with standard (RTDF) mix; (D) Treatment 4: fertilized, planted with local (U of SK) mix. The PHC data were grouped into four PHC-fractions (based on equivalent C number): F1 (C6–10), F2 (C10–16), F3 (C16–34), F4 (C34–50).



Changes in PHC concentration with time exhibited a similar pattern for all four treatments, though reductions in the F3 fraction generally occurred more rapidly in the planted treatments. Overall, however, data analysis indicated that (i) there were no significant differences between the site-specific (USASK) and the standard (RTDF) plant mixes and (ii) there were no significant differences between the planted and unplanted treatments.

Reductions in PHC concentrations were generally greater in the plots amended with fertilizer and compost. However, because of the high degree of spatial variability associated with the PHC determinations, statistical significance was not observed. Concentrations of PAHs in the surface soils also decreased significantly after four years (Table 4), and were well below the SQG upon completion of the study. Naphthalene concentrations in the sub-surface decreased by an average of about 45% (to an average of 1.3 mg kg^{-1}), but were still about 30% greater than the SQG (0.1 mg kg^{-1}).

Significant changes in both electrical conductivity (EC) and sodium adsorption ratio (SAR) were observed, with site-averaged decreases of 34% and 71%, respectively. In addition, significant treatment effects observed with soils from the amended soils yielding EC and SAR values that were 3–4 times lower than soils from the unamended plots. Plant

effects on salinity were not observed in the surface 15-cm, but would have been expected to occur in the sub-surface[5]. That is, root penetration into the subsurface (which was visually observed) would be expected to increase water infiltration–leaching salts from the deeper layers.

Trace metals were not determined at the final sampling, reflecting the fact that they did not exceed the SQGs at the start of the study. Analysis of a subset of the above-ground biomass from the planted treatments failed to detect any PHCs in the plant tissue. This is in keeping with the vast majority of the published literature, which indicates that plant uptake of PHCs, and most PAHs, during phytoremediation are negligible. As a result, a detailed analysis of the plant tissues was not undertaken.

Conclusions

Significant reductions in TPH and the CCME PHC-fractions were observed during the four years of field trials at the Talisman Energy site near Carlyle, AB. Reductions in the F3 (C16–C34) fraction, benzene, toluene, ethylbenzene, naphthalene, and phenanthrene brought the final concentrations of these compounds in the surface soil to levels below those identified in the *Canadian Soil Quality Guidelines* as needing further remedial action. Despite significant reductions in the F3 fraction (*ca*. 74%) and naphthalene (*ca*. 45%) in the sub-surface soils, the final concentrations of these compounds still exceeded the SQG for a fine-grained agricultural soil. Total reductions in PHC concentrations in the planted and unplanted treatments were generally similar, though the degradation kinetics appear to be more favourable in the planted treatments. As well, plant establishment provides additional benefits of enhanced soil stabilization and erosion and runoff control.

[5] Due to a communication error, sub-surface soils from the planted treatments were not analyzed. These analyses will be conducted as soon as possible, and the results appended to the report.