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Lucas Odom

Joel Gerard Burken
Missouri University of Science and Technology, burken@mst.edu

Lee Newman

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Distribution and Accumulation of Trichloroethylene and Trichloroacetic Acid in Hybrid Poplars

Lucas Odom[1]; Joel Burken, P.E., M.ASCE[2]; and Lee Newman[3]

Abstract: Hybrid poplar trees are known to take up trichloroethylene (TCE) into above ground tissues, where it degrades into the metabolites trichloroacetic acid (TCAA), dichloroacetic acid, and trichloroethanol and where parent chlorinated solvents volatilize to the atmosphere. Based on this knowledge, numerous phytoremediation applications have been implemented for TCE and other chlorinated solvents. Sampling of plant tissues has proven effective for phytoforensic and phytoscreening applications in assessing sites and evaluating the efficacy of phytoremediation. However, little is known about the appropriate exposure times and sampling locations required to obtain an accurate assessment of TCE and metabolites. In this study, hybrid poplars were dosed under greenhouse conditions for times ranging from one hour to 29 weeks. No increasing accumulation of TCE occurred in the stems over the time periods; however, concentrations decreased with increasing stem height in all cases. The concentration of TCAA throughout the plants' leaves fluctuated along the stem rather than following a decreasing pattern with height. However, as a result of its nonvolatile characteristics, TCAA did accumulate to higher concentrations in the leaves over the given time periods. These results revealed that sampling TCE in the lower stem/trunk was ideal, but TCAA concentrations varied across locations; thus, sampling of multiple leaves is appropriate for an accurate determination of accumulative contaminant uptake. Used together, these methods offer a novel monitoring tool, which is needed because phytoremediation does not offer traditional monitoring means such as treated effluent to assess fate and efficacy. Knowledge resulting from this research can improve monitoring and reduce long-term monitoring costs for chlorinated solvent sites. **DOI: 10.1061/(ASCE)EE.1943-7870.0000703.** © *2013 American Society of Civil Engineers.*

CE Database subject headings: Acids; Trees; TCE.

Author keywords: Phytoremediation; Trichloroethylene; Distribution; Monitoring.

Introduction

Trichloroethylene (TCE) is a common contaminant of soils and groundwater because of accidental spills, deliberate dumping into the environment, and use as an industrial degreaser (Halsey et al. 2005). Because groundwater is the largest reservoir of freshwater, with stored volumes of 50 times greater than the amount of surface freshwater (Bayer and Finkel 2006), and because groundwater is often utilized with minimal or no treatment, efforts are needed to efficiently and effectively clean up existing problems such as TCE in groundwater.

TCE can exist as a soil or groundwater contaminant, migrating deep into aquifers as a dense nonaqueous phase liquid (DNAPL) (Cho et al. 2005). TCE is chemically stable and often biologically recalcitrant in aquifers, with half-lives measured in years, and the NAPL provides a long-term secondary source of contamination due to limited solubility and low mass transfer rates (Bayer and Finkel 2006). These properties and ubiquitous use have led to TCE becoming the one of the most prevalent groundwater contaminants in the US, and drinking water is the leading human exposure route (EPA 2007). Additionally, the classification on TCE has recently been changed from a possible human carcinogen; TCE is now listed as a human carcinogen (EPA 2011).

Research has shown that hybrid poplars and other deciduous trees such as sycamore and sweet gum, and evergreens such as pine and Leland cypress (Strycharz and Newman 2009a, b) have the ability to transport TCE from the groundwater into above-ground portions of the plant (Newman et al. 1997). Following uptake, TCE can be degraded to the metabolites trichloroacetic acid (TCAA), dichloroacetic acid (DCAA), and trichloroethanol (TCOH), and also volatilizes to the surrounding atmosphere (Newman et al. 1997; Struckhoff et al. 2005). In the atmosphere, TCE and other chlorinated solvents have half-lives measured in as low as five days (EPA 2007). Phytoremediation offers an inexpensive and efficient TCE remediation tool that regulators and the industry have initiated at numerous sites (Cho et al. 2005). Additionally, phytoremediation is more publically embraced and aesthetically pleasing to a community than excavation or to pump-and-treat operations, which are reliant on energy-consuming water treatment facilities on site. Phytoremediation also adheres to the green remediation emphasis of the EPA and the remediation field. In addition, phytoremediation offers ancillary ecosystem service benefits of measurable value and health benefits (Holzman 2012).

Because TCE has multiple fates in phytoremediation systems, lingering questions exist regarding the ultimate fate and distribution of TCE and its metabolites within tree tissues. With better understanding of sampling locations and exposure periods for optimal tissue concentrations, more efficient site plant sampling can

[1]Environmental Health Sciences, Univ. of South Carolina, Columbia, SC 29208; presently at, Shealy Environmental, West Columbia, SC 2917.

[2]Associate Chair and Professor, Dep. of Civil, Architectural and Environmental Engineering, Missouri Univ. of Science and Technology, Rolla, MO 65409-0030.

[3]Associate Professor, Environmental and Forest Biology, SUNY College of Environmental Science and Forestry, Syracuse, NY 13210 (corresponding author). E-mail: lanewman@esf.edu

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accurately determine and quantify the exposure and efficacy of the phytoremediation systems. The purpose of this study was to determine whether the time since TCE exposure commences impacts the concentration of TCE and metabolites within the plant, and how sampling location on the plant might also influence concentration. Comparing data with existing fate models can provide improved knowledge regarding the interactions of the multiple fates. With better understanding of fate and kinetics, improved plant sampling methods can be developed and data can be interpreted correctly to accurately quantify the efficacy of the phytoremediation systems.

Materials and Methods

Growing, Dosing, and Harvesting Plants

Hardwood cuttings (25 cm in length) of hybrid poplar clone 0P-367 (*Populus deltoides* x *P. nigra*) were used for all studies. Five control plants and five plants dosed with the TCE solution were set up for each sampling time point. The poplars were grown in 2-L beakers, which were filled with Landscaper's Choice topsoil underlain with 25 mm (1 in.) of pea gravel to prevent soil saturation and improve the distribution of feedwater. A glass watering tube was run to the bottom of the beaker and into the pea gravel to minimize the volatilization of TCE and mimic groundwater exposure. Beakers were wrapped in foil to prevent photodegradation of TCE and to prevent algae growth (Fig. 1). The plants were grown until roots were developed enough to visibly penetrate into the pea gravel layer. The greenhouse was set with daytime temperatures of 27°C and nighttime temperatures of 17°C, and daylight was extended to 14 h each day using 400-W Phillips sodium vapor greenhouse lights.

Plants were dosed three times per week with a 100 ppm TCE solution. The solution was prepared immediately before application from a saturated stock solution, and this solution was added to plant root zones via the watering tube. This concentration was selected as one that did not have an impact on the growth of this poplar line when grown in soils, and also resulted in rapid detection of the TCE and metabolites in the plants. The amount of added TCE solution depended on the individual plant's duration of dosing (Table 1). Short-term exposure plants were harvested 1, 5, 10, and 24 h after the initial dose. Long-term exposed plants were harvested 1, 2, 3, 5, and 29 weeks after the initial dose, again with dosing taking place three times per week during the listed periods. Control plants were placed upwind of the dosed plants with regard to ventilation fans and were grown under the same conditions but were not dosed with TCE.

The plants were harvested in 5–10-cm sections. Starting from the top of the plants, the first section of leaves was removed, flash frozen in liquid nitrogen, and stored in volatile organic analysis (VOA) vials on dry ice. This process was continued down the stem until all of the leaves were removed. The section of the stem that corresponded with the section of leaves was clipped and flash frozen in liquid nitrogen, placed in individual VOA vials, and stored on dry ice. These steps were continued for the entire plant. Once harvesting was complete, the samples were stored at −80°C until extraction. A minimum of three replicate plants was used for each time point. This research does not attempt to close a mass balance, which has been completed in earlier work (Newman et al. 1997, 1999; Burken and Schnoor 1998), but rather attempts to assess TCE and metabolite fate in tissues.

Extractions and Analysis of TCE

Stem samples were extracted and analyzed for TCE. Each stem section was individually broken into small splinters by using liquid nitrogen to freeze the tissue, pliers to crush the stem, and a mortar and pestle to grind the tissue in liquid nitrogen, which prevented the loss of TCE from the tissues. The ground sample was placed into 2 mL of 1 N H_2SO_4/10% NaCl solution in a glass vial with a Teflon lined cap to prevent loss of TCE during extraction. Ten milliliters of methyl *tert*-butyl ether (MTBE) was added and the mixture was

Fig. 1. Hybrid poplar in a 2-L beaker, showing foil wrapping and watering tube along the side of the beaker

Table 1. Data for Total TCE Mass, Water Added to Reactors, and New Stem Diameters of Hybrid Poplars for Each Dosing Period

Dosing period	TCE received (mg)	Stem diameter (mm)	Water uptake (mL)
1 h	22	4.7 ± 0.3	3.5 ± 1
5 h	22	4.7 ± 0.2	46.4 ± 8.3
10 h	22	4.4 ± 0.2	85.2 ± 3.8
24 h	22	4.4 ± 0.6	86.4 ± 10.7
1 week	66	4.7 ± 0.7	—
2 week	132	4.7 ± 0.8	—
3 week	198	4.7 ± 0.9	—
5 week	330	7.5 ± 0.10	—
29 week	1,914	10.8 ± 0.3	—

Note: Data presented as ± one SD.

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shaken for 10 min at 300 rpm. The samples were allowed to rest for 10 min and the MTBE layer was decanted onto 2 g of Na_2SO_4 for 45 min to remove any residual water. The MTBE was pipetted into a 2 mL capillary gas chromatography vial with an internal standard of 2 μL of ethylene dibromide (50 μL/100 mL MTBE).

TCE extracts were run on a Perkin Elmer AutoSystem gas chromatograph (GC) with a built-in autosampler, a Supelco VOLCOL column (60 × 0.25 m × 1.25 μm film thickness) with helium as the carrier gas. Each sample was run for 58 min at 140°C with an injection volume of 1.0 μL and a flow rate of 20 mL/ min. An electron capture detector (ECD) was used at a temperature of 340°C.

Extractions and Analysis of Metabolites

Each leaf sample was extracted and analyzed for TCAA by using a modification of USEPA Method 552 (Hodgeson et al. 1990). Each leaf sample was ground into a powder with liquid nitrogen to prevent loss of metabolites, mixed with 20 mL of nanopure water, and shaken for 10 min at 300 rpm in a glass vial with a Teflon lined cap. After shaking, 3 mL of concentrated H_2SO_4, immediately followed by 9 g of Na_2SO_4/1 g of $CuSO_4$ and 7 mL of MTBE, were added to the leaves. Again, this was shaken for 10 min at 300 rpm and allowed to settle for 5 min. The MTBE layer was transferred to a 40 mL VOA vial, where 6 mL of 10% H_2SO_4 in methanol was added to the MTBE extract. The vial was placed in a 50°C water bath for 1 h, cooled at 4°C for 10 min, and 15 mL of a $CuSO_4/Na_2SO_4$ solution (50 and 100 g/L, respectively) was added. The mixture was shaken again for 10 min at 300 rpm and allowed to rest for 5 min. The MTBE layer was transferred to a 2 mL capillary GC vial with 2 μL of ethylene dibromide (50 μL/100 mL MTBE).

Extracts were run on the same GC system as the TCE extracts with only minor adjustments. The run time was extended to 2 h at 100°C and the flow rate was decreased to 10 mL/ min. Method and sample spike recoveries were generally in the range of 70–85%, depending on the extraction run.

TCE Modeling

The data for TCE in the stems was plotted and compared to the output of a previously developed TCE fate model (Ma and Burken 2004). The model equation [Eq. (1)] was used to calculate the concentration (C) in the stem at a height (z) based upon the ground-level concentration and the diffusional losses as the volatile compound ascends up the transpiration pathway. The conceptual basis of the model is the radial loss from chlorinate solvents from the active xylem tissues transporting the volatile compounds:

$$C = C_0 e^{-[(Ra/R-Ra)]\times(2Dr\pi/Q)z} \quad (1)$$

The descriptions of the variables are in Table 2. This model predicts that as the distance up the stem increases, the concentration of TCE decreases exponentially. The model has been applied to various other studies (Gopalakrishnan et al. 2007; Yin et al. 2011) and used to validate previous conceptual models on contaminant transport in trees (Baduru et al. 2008). In the work by Yin and colleagues (2011), the analytic solution for the fundamental model was applied to data for polycyclic aromatic hydrocarbons (PAHs), validating the model for a range of compounds relative to vertical transport and volatilization.

Statistical Analysis

All statistics were determined by using *SAS version 9.1.3*. One-way ANOVA was used to compare differences between the treatment groups. Pearson's correlation coefficient was used to determine the significance of the relationship between TCE and TCAA concentrations.

Table 2. Descriptions of Variables Given by Previously Developed Model

Variable	Description	Value applied
C	Concentration (mg/L)	
C_0	TCE concentration in transpiration stream at height zero (mg/L)	Actual concentration, determined analytically
Ra	Effective diffusion path (cm)	0.85 R
R	Radius (cm)	Actual radius
Dr	Diffusivity in radical direction (cm^2/s)	1.24×10^{-7}
Q	Transpiration rate (cm^3/s)	Model input (Table 1)
z	Height up stem (cm)	Model input (x-axis of Fig. 6)

Note: Data from Ma and Burken (2004).

Results

TCE in Stems: Duration of Dosing

The largest factor that contributed to TCE concentration in the stems was the amount of time dosing took place before sampling. When plants were dosed once, 1 h before sampling, no TCE was found in any of the stems. However, TCE concentrations peaked when dosing took place 5 h before harvesting. TCE concentrations decreased 10 h after exposure and decreased even further 24 h after dosing once with TCE (Fig. 2). The decrease in plant concentrations over this period may be attributable to metabolism or equilibration as the parent TCE reaches sorption equilibrium with soil and uptake by plant tissues. The same effect was found in plants dosed for multiple weeks. When plants dosed for 5 weeks were harvested 5 h after dosing, concentrations of TCE were higher in the lower portions of the stem than plants dosed for 29 weeks (Fig. 3), which were harvested 24 h after the last TCE dosing. However, overall stem concentrations were more uniform in plants dosed for longer time periods.

The results of this study show that the length of exposure to TCE plays a limited role in determining the concentration of TCE in the stems of hybrid poplars. Plants dosed for only one week had similar TCE concentrations to plants dosed for 3 weeks (Fig. 4). Furthermore, the concentration of TCE in plants dosed once for 5 h was not statistically different ($\alpha = 0.05$) from plants dosed for 29 weeks but harvested 24 h after the final dosing event. However, plants dosed for 29 weeks were significantly but not dramatically higher than plants dosed for 1, 2, and 3 weeks when all plants were harvested 24 h after dosing.

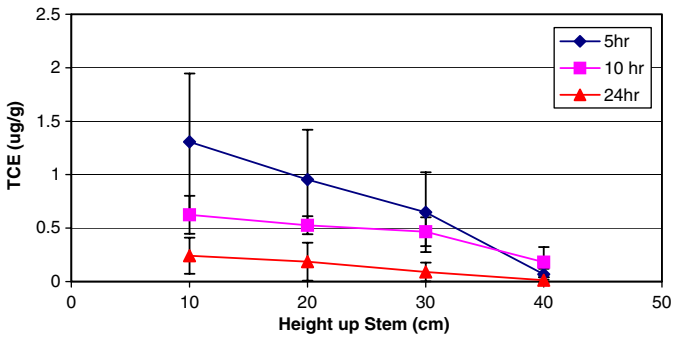

Fig. 2. Distribution of TCE in hybrid poplars compared to height along stem (TCE was measured in μg/g and height was measured in cm; error bars are one SD from the mean of replica plants)

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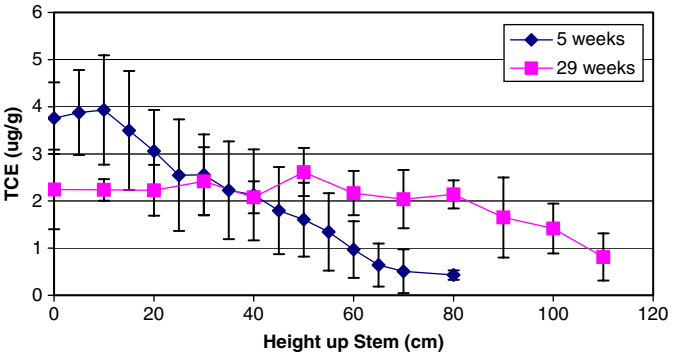

Fig. 3. Comparison of TCE distribution between plants exposed for 5 and 29 weeks as height up stem increases (TCE was measured in μg/g and height was measured in cm; error bars are one SD from the mean of replica plants)

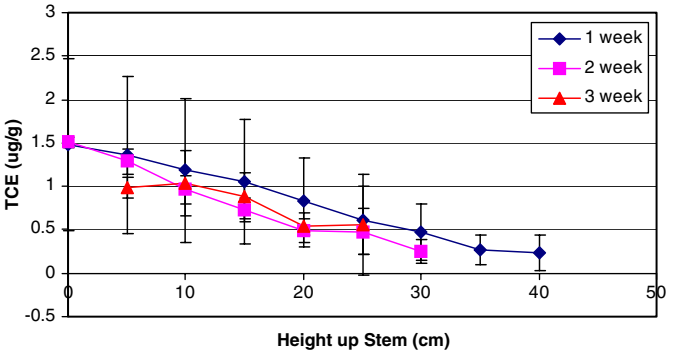

Fig. 4. Comparison of TCE distribution in hybrid poplars dosed for 1, 2, and 3 weeks as height up stem increases (TCE was measured in μg/g and height was measured in cm; error bars are one SD from the mean of replica plants)

TCE in Stems: Localization

For every plant in the study, the highest concentrations of TCE were found in the lowest section of the stem. As height increased, the concentration of TCE in the stem uniformly decreased (Figs. 2–4). Concentrations of TCE varied in the base of the stems for each time period, but concentrations were similar for each time period at the tops of the stems. The decrease with height is expected, as a result of volatilization along the transpiration path and degradation to metabolites within the plant tissues.

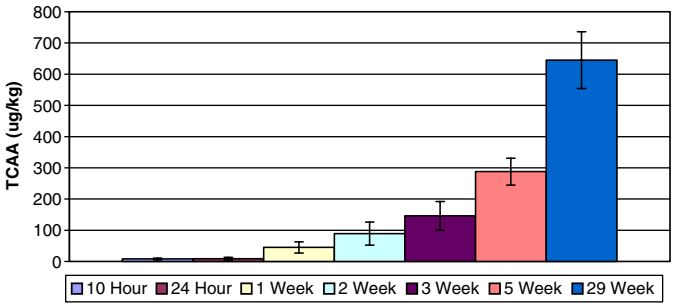

Fig. 5. TCAA concentrations in all leaves of hybrid poplars exposed to TCE (TCE was measured in μg/g; error bars are one SD from the mean of replica plants)

TCAA in Leaves

This study focused primarily on TCAA in the leaves as the predominant metabolite of TCE exposure in hybrid poplars. DCAA can be found in hybrid poplar leaves exposed to TCE, but is also present in background levels in unexposed plants, and levels do not increase as dramatically as for TCAA following TCE exposure.

The exposure time of hybrid poplars to TCE did have an impact on the concentration of TCAA in the leaves. As exposure time increased, the levels of TCAA in the leaves increased for most time periods. Plants exposed to TCE for only 1 h did not have any detectable amounts of TCAA. Plants dosed for 5 h showed small amounts of TCAA, but the concentrations were not above quantification limits. TCAA was not found in any of the control plants and growth was not notably different from dosed plants.

No statistically significant increase in TCAA levels was observed with time increases between 10 and 24 h (Fig. 5). After one week, the overall average TCAA concentration in the leaves increased with the length of TCE exposure although the differences were not statistically significant between one and two weeks, and between two and three weeks. Hybrid poplars dosed for 29 weeks had much higher TCAA concentrations than those dosed for any other time period. Because TCAA is a relatively nonvolatile metabolite, the increase in TCAA concentration with increased accumulative TCE exposure is expected and reveals the continued degradation of TCE.

Unlike TCE, TCAA levels did not correlate with the height of the stem. Rather, TCAA concentrations fluctuated throughout the plant (Table 3). Fluctuations were more considerable in plants dosed for longer time periods. In plants dosed for 29 weeks, the

Table 3. Concentrations of TCAA in Leaves of Hybrid Poplars with Increases in Height

	Height up stem (cm)								
Dose time	0	10	20	30	40	50	60	70	80
	Concentration of TCAA (μg/kg)								
1 h	0	0	0	0	0	—	—	—	—
5 h	TLQ	TLQ	TLQ	TLQ	TLQ	—	—	—	—
10 h	—	2.7 ± 3.7	4.2 ± 5.9	8.8 ± 6.4	10.1 ± 6	—	—	—	—
24 h	—	6.5 ± 5.9	10.2 ± 1.8	9.3 ± 5.8	8.7 ± 8.2	—	—	—	—
1 week	59.8 ± 16	25 ± 21.9	71 ± 42	17.7 ± 18	24.1 ± 13	—	—	—	—
2 week	147 ± 91	80 ± 61.2	93.3 ± 67	136.9	136.6	—	—	—	—
3 week	135 ± 100	99.1 ± 24	78 ± 12.4	170.0	—	—	—	—	—
5 week	392 ± 113	320 ± 36	329 ± 43	229 ± 60	329 ± 93	270 ± 108	279 ± 143	154 ± 82	130.4
29 week	656 ± 219	628.0	661.3	718 ± 60	556 ± 275	717 ± 283	664 ± 244	769 ± 133	428 ± 83

Note: TCAA concentrations were recorded (in μg/kg) and heights were measured in cm. Concentrations are written with ± one SD. Samples are indicated with identified TCAA, but at concentrations too low to quantify (TLQ). Plants exposed for 1 to 24 h were dosed once, with time indicating hours after exposure that the sample was collected.

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fluctuations were as large as 500 μg of TCAA/kg leaf tissue in leaves collected from harvested stem sections.

TCE/TCAA Relationship

The accumulation of TCAA in the leaves appears to be independent of the concentration of TCE in the stem at any specific time. When comparing the TCAA levels in the leaves harvested from a given stem section and the TCE in that stem section, no relationship could be found ($R^2 = 0.018$). Thus, high levels of TCE at a given stem height do not indicate a corresponding high concentration of TCAA at that same height.

Discussion

TCE in Stems

Plants dosed for only 1 h did not have any measurable amounts of TCE or TCAA within their stems or leaves, respectively, which is attributed to the limited transport time to move TCE from the roots to the stem. In the 1-h time period, only 3.5 mL, on average, of water was taken up by the plants (Table 1).

Plants dosed once and exposed to TCE for 5 h had a larger amount of TCE concentrations in the stem than plants that were only dosed with TCE once. Plants dosed once and exposed for 10 and 24 h took up, on average, 86 mL of water, which is double the amount that the plants exposed for 5 h took up. Lower TCE concentrations in the 10 and 24 h plants are probably attributable to the volatile nature of TCE. This is probably two factors at work: diffusion of the TCE out of plant stems, and a lower concentration of TCE in the transpiration stream coming up from the roots due to TCE volatilizing from the solution at the bottom of the beakers and into the overlaying soil, where sorption limits exposure concentration and plant uptake.

Studies have been developed to model the transport and resulting concentrations of VOCs in plant tissues. The research has shown that as the distance up the stem increases, the concentration of TCE in the stem decreases. Likewise, as the distance up the root increases, the concentration of TCE in the root decreases (Newman et al. 1997; Ma and Burken 2003). The trend of decreasing TCE concentrations with height in this study fit well with the applied model based upon volatilization from the stem (Fig. 6). However, plotting residuals revealed clear model biases, low at the base of the plant and high at top of the plant. For this single application of the model to laboratory scale data, the change in the stem radius is a likely cause of the underprediction of the volatility losses, because the stem decreases in radius with height, which the current model is not able to adjust at the different heights. The model was also developed for lignified xylem tissues, not new growth stem as applied in this case. The fit of model data here to a new system indicated the robust nature of the mechanistic model, applied to different tissues and multiple scales, from ≈4-mm-diameter new growth stems to >20-cm trees over 10 m high (Ma and Burken 2004).

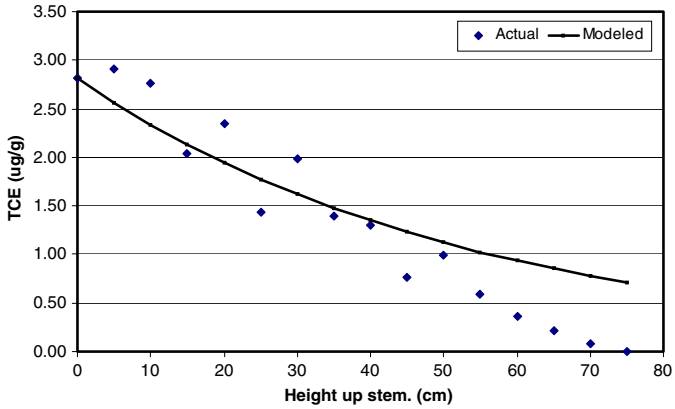

Fig. 6. Comparison of model output to actual concentrations of TCE in stems: actual values are samples from an individual tree (TCE concentrations were measured in μg/g)

The importance of diffusion path length was shown in the initial model development, revealing that the stem diameter plays a large role in the distribution of volatile compounds with elevation (Ma and Burken 2004). The results of this study support this, because plants dosed for 29 weeks had greater diameters than all other plants (Table 1). These plants also had greater TCE concentrations with elevation than all other plants sampled 24 h after their last dosing. The importance of diffusion path length was also confirmed in recent research with this fundamental model on PAHs in field studies (Yin et al. 2011).

Caution must be used when translating findings from greenhouse studies and models to fully grown trees in the field. Differences in tissue chemistry, bark thickness, distance from xylem to trunk edge, and increased efficiency of metabolic processes in the field will all have an impact on TCE transport and levels in the plants. The difficulty in conducting these types of studies with field plants is primarily due to uncertainties about actual contaminant concentration exposure and varying environmental conditions.

TCAA in Leaves

TCAA accumulated in leaves over exposure time as a result of its nonvolatile nature. Also, the leaves may act as a depository for TCAA until further metabolism or removal occurs (Schroder et al. 2003). In this study, the equilibrium or peak concentration of TCAA was likely not reached, but the increasing trend and deposition of TCAA in leaves was clearly demonstrated. To better determine the steady-state concentrations, more time periods need to be tested to determine the kinetic parameters and to establish when TCAA concentrations no longer increase. In previous research, TCAA concentrations in hybrid poplars were found to range from 1 to 7 μg/g after 31 weeks of TCE exposure (Newman et al. 1997). These concentrations are only slightly higher than the concentrations found in the hybrid poplars dosed for 29 weeks in this study. The results of the laboratory studies indicate that TCAA concentrations in leaves can indicate cumulative exposure to TCE, whereas sampling of the stem for parent TCE indicates the current concentration of groundwater exposure.

This hypothesis was supported in a full-scale field study as TCAA concentrations in leaves averaged 14.5 μg/g in two-year-old hybrid poplars exposed to TCE for 16 weeks (Newman et al. 1999). In the full scale study, recorded TCAA concentrations were approximately three times higher than any TCAA concentrations found in this greenhouse study. However, the grown field plants had taken up approximately 20 times more TCE than any plant dosed in this study.

Conclusions

In assessing contaminant fate in phytoremediation efforts, novel methods are needed that can determine treatment efficacy. Plant sampling methods can now provide insight into contaminant removal rates from parent compound and metabolite concentrations. As with any developing methods and approaches, advanced knowledge is needed to best interpret novel, atypical data. When

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sampling leaves either in the field or the greenhouse, the location on the stem may not be as important when testing for metabolites such as TCAA as it is when testing for parent compounds such as TCE. However, sampling multiple leaves to account for variations in metabolite levels is important.

When sampling small scale stems for TCE during greenhouse studies, 5 h appears to be sufficient for detecting uptake. The exposure to TCE for more than one week did not result in a significant difference in the concentration of TCE in the stem in this study. The highest concentrations of TCE are found at the lowest portion of the stem, independent of the length of TCE exposure. The rapid responses to changes in concentration indicate that the concentration of the TCE is a temporally sensitive measure and can vary over medium to short periods after exposure changes (Vroblesky et al. 2004); recent research has indicated considerable seasonal variations.

For field studies, sampling multiple leaves from various trees and locations on a given tree can average out spatial differences. Additionally, sampling well into the growing season should result in the highest TCAA concentrations, in addition to stable TCE concentrations in the stems and trunks. For TCE stem sampling, sampling midday to early afternoon when transpiration has been active for at least 5 h and is at its peak appears to give the best chance for the highest stem or trunk concentrations.

Better knowledge and improved methods can alleviate some of the ambiguity related to contaminant fate and the assessment of efficacy in phytoremediation applications. This can also be useful information sites where sampling native plants can be used in phytoscreening and phytoforensics (Burken et al. 2011). Evaluating the ratios of parent compounds to metabolites offers new insights into the relationship of current exposure concentration and cumulative contaminant exposure over a growing season.

References

Baduru, K. K., Trapp, S. T., and Burken, J. G. (2008). "Direct measurement of VOC diffusivities in tree tissues: Impacts on tree-based phytoremediation." *Environ. Sci. Technol.*, 42(4), 1268–1275.

Bayer, P., and Finkel, M. (2006). "Life cycle assessment of active and passive groundwater remediation technologies." *J. Contam. Hydrol.*, 83(3), 171–199.

Burken, J. G., and Schnoor, J. L. (1998). "Predictive relationships for uptake of organic contaminants by hybrid poplar trees." *Environ. Sci. Technol.*, 32(21), 3379–3385.

Burken, J. G., Vroblesky, D. A., and Balouet, J. C. (2011). "Phytoforensics, dendrochemistry, and phytoscreening: New green tools for delineating contaminants from past and present." *Environ. Sci. Technol.*, 45(15), 6218–6226.

Cho, C., Sung, K., Coapcioglu, M. Y., and Drew, M. (2005). "Influence of water content and plants on the dissipation of chlorinated volatile organic compounds in soil." *Water Air Soil Pollut.*, 167(1–4), 259–271.

EPA. (2007). "Trichloroethylene (TCE)." USEPA Toxicity and Exposure Assessment for Children's Health (TEACH) summary, ⟨http://www.epa.gov/teach/chem_summ/TCE_summary.pdf⟩ (2008).

EPA. (2011). "Toxicological review of trichloroethylene." *EPA/635/R-09/011F*, Washington, DC.

Gopalakrishnan, G., Negri, M. C., Minsker, B. S., and Werth, C. J. (2007). "Monitoring subsurface contamination using tree branches." *Ground Water Monit. Rem.*, 27(1), 65–74.

Halsey, K. H., Sayavedra-Soto, L. A., Bottomley, P. J., and Arp, D. J. (2005). "Trichloroethylene degradation by butane-oxidizing bacteria causes a spectrum of toxic effects." *Appl. Microbiol. Biotechnol.*, 68(6), 794–801.

Hodgeson, J. W., Collins, J., and Barth, R. E. (1990). "Method 552.0, Determination of haloacetic acids and dalapon in drinking water by liquid-liquid microextraction, derivatization, and gas chromatography with electron capture detection." Technical Support Center Document, Office of Ground Water and Drinking Water, U. S. Environmental Protection Agency.

Holzman, D. C. (2012). "Accounting for nature's benefits: The dollar value of ecosystem services." *Environ. Health Perspect.*, 120(4), a152–a157.

Ma, X., and Burken, J. G. (2003). "TCE diffusion to the atmosphere in phytoremediation applications." *Environ. Sci. Technol.*, 37(11), 2534–2539.

Ma, X., and Burken, J. G. (2004). "Modeling of TCE diffusion to the atmosphere and distribution in plant stems." *Environ. Sci. Technol.*, 38(17), 4580–4586.

Newman, L. A., et al. (1997). "Uptake and biotransformation of trichloroethylene by hybrid poplars." *Environ. Sci. Technol.*, 31(4), 1062–1067.

Newman, L. A., et al. (1999). "Remediation of trichloroethylene in an artificial aquifer with trees: A controlled field study." *Environ. Sci. Technol.*, 33(13), 2257–2265.

SAS version 9.1.3 [Computer software]. SAS Institute, Cary, NC.

Schroder, P., Matucha, M., Forzcek, S. T., Uhlirova, H., Fuksova, K., and Albrechtova, J. (2003). "Uptake, translocation and fate of trichloroacetic acid in a Norway spruce/soil system." *Chemosphere*, 52(2), 437–442.

Struckhoff, G., Burken, J. G., and Schumacher, J. (2005). "Phytoremediation of vadose zone VOCs." *Environ. Sci. Technol.*, 39(6), 1563–1568.

Strycharz, S., and Newman, L. A. (2009a). "Use of native plants for the remediation of trichloroethylene I: Deciduous trees." *Int. J. Phytoremed.*, 11(2), 150–170.

Strycharz, S., and Newman, L. A. (2009b). "Use of native plants for the remediation of trichloroethylene II: Coniferous trees." *Int. J. Phytoremed.*, 11(2), 171–186.

Vroblesky, D. A., Clinton, B. D., Vose, J. M., Casey, C. C., Harvey, G. J., and Bradley, P. M. (2004). "Groundwater chlorinated ethenes in tree trunks—Case studies, influence of recharge, and potential degradation mechanism." *Ground Water Monit. Rem.*, 24(3), 124–138.

Yin, H., Tan, Q., Chen, Y., Lv, G., He, D., and Hou, X. (2011). "Polycyclic aromatic hydrocarbons (PAHs) pollution recorded in annual rings of gingko (*Gingko biloba* L.): Translocation, radial diffusion, degradation and modeling." *Microchem. J.*, 97(2), 131–137.