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METAL UPTAKE AND ALLOCATION IN TREES GROWN ON CONTAMINATED LAND: IMPLICATIONS FOR BIOMASS PRODUCTION

Michael W.H. Evangelou,[1] Brett H. Robinson,[2] Madeleine S. Günthardt-Goerg,[3] and Rainer Schulin[1]
[1]*Institute of Terrestrial Ecosystems, ETH Zürich, Zürich, Switzerland*
[2]*Agriculture and Life Sciences, Lincoln University, Lincoln, Canterbury, New Zealand*
[3]*Swiss Federal Research Institute for Forest, Snow and Landscape (WSL), Birmensdorf, Switzerland*

Phytostabilization aims to reduce environmental and health risks arising from contaminated soil. To be economically attractive, plants used for phytostabilization should produce valuable biomass. This study investigated the biomass production and metal allocation to foliage and wood of willow (**Salix viminalis L.*****), poplar (*****Populus monviso*****), birch (*****Betula pendula*****), and oak (*****Quercus robur*****) on five different soils contaminated with trace elements (TE), with varying high concentrations of Cu, Zn, Cd, and Pb as well as an uncontaminated control soil. In the treatment soils, the biomass was reduced in all species except oak. There was a significant negative correlation between biomass and foliar Cd and Zn concentrations, reaching up to 15 mg Cd kg^{-1} and 2000 mg Zn kg^{-1} in willow leaves. Lead was the only TE with higher wood than foliage concentrations. The highest Pb accumulation occurred in birch with up to 135 mg kg^{-1} in wood and 78 mg kg^{-1} in foliage. Birch could be suitable for phytostabilization of soils with high Cd and Zn but low Pb concentrations, while poplars and willows could be used to stabilise soils with high Cu and Pb and low Zn and Cd concentrations.***

KEY WORDS: bioenergy, heavy metals, birch, phytostabilization, poplar, willow

INTRODUCTION

Trace elements (TE) occur naturally in the Earth's crust, but human activities have altered their geochemical cycles. Soil contamination with TE results from land application of biosolids, waste incineration, agricultural use of low quality fertilisers and application of TE-based pesticides, mining, and processing of TE ores, industrial and traffic emissions, weathering of buildings, military activities and others (Kozlov, Haukioja, Bakhtiarov *et al.* 2000; Sterckeman, Douay, Proix *et al.* 2002; Sebastiani, Scebba and Tognetti 2004; Walter, Martinez and Cala 2006). Soil pollution by TE is responsible for losses in soil fertility, food contamination, ecotoxic effects and risks to human health.

Address correspondence to Michael W.H. Evangelou, Institute of Terrestrial Ecosystems, ETH Zürich, Universitätstrasse 16, CH-8092, Zürich, Switzerland. E-mail: michael.evangelou@env.ethz.ch

Landfilling of TE-contaminated soil not only results in the loss of otherwise fertile soil, but also is expensive. Clean-up is often not possible for economic and/or ecological reasons. Thus, keeping the contamination in place under control, from being dispersed into the environment and preventing it from reaching food chains are the only treatment options. This is the goal of phytostabilization. In this approach a vegetation cover is used to reduce leaching by extracting soil water via transpiration and reducing soil erosion (Robinson *et al.* 2006). The effectiveness of vegetation in controlling leaching depends on the soil properties as well as on the climate. Deep rooted plant species are generally more suitable for phytostabilization than shallow rooted species, especially in dryer climates or dry periods, because shallow rooted species have less access to water and are, therefore, more likely to suffer from droughts (Vogeler *et al.* 2001). Thus, a permanent cover would not be guaranteed and a dispersion of contaminated soil via wind or water erosion would become possible. Besides acting as umbrellas and preventing some 15% of rainfall from reaching the ground by evaporating intercepted precipitation, tree canopies also reduce evaporation from the ground and thus keep the forest floor moist (Robinson *et al.* 2009). Additionally, by extracting soil moisture, roots help maintain aerobic conditions in the vadose zone, preventing reduction of redox-sensitive TE, which are often more toxic and more mobile than oxidized species of the same TE. Plants add organic matter to the soil in the form of litter and root exudates which bind TE (Pulford and Watson 2003; Robinson *et al.* 2009).

However, phytostabilization is only economically attractive if it also enhances the value of the land through ecological benefits or the production of nonedible biomass for commercial products such as bioenergy or timber. For commercial biomass production on contaminated soil, tree species are needed which have, 1) a high biomass production, 2) a high root biomass to stabilize the soil and the contaminants, 3) a high tolerance combined with low TE uptake, especially in the foliage, as leaves can enter the food chain, and 4) easy handling (e.g., short rotation coppice) (Perronnet *et al.* 2000). Although, there are many studies that have investigated phytostabilization and phytoextraction of TE contaminated soils with willow and poplar (Pulford and Watson 2003; Mertens *et al.* 2004; Sebastiani, Scebba and Tognetti 2004; Dickinson and Pulford 2005; Brunner *et al.* 2008) there are still several issues that have remained unanswered. It is not clear how naturally contaminated soil and soil properties affect TE uptake for specific tree species. Furthermore, the implications of TE uptake for fuel production from biomass of TE contaminated are unknown. Finally, the role of elements such as Sb which have been neglected but which are important due to their toxicity and their mobility (Johnson *et al.* 2005; Sorvari, Antikainen, and Pyy 2006) has to be addressed.

In the present study, we investigated the possibility of combining phytostabilization and bioenergy production by growing trees on TE contaminated soils. For this purpose we compared the TE accumulation and biomass production of four tree species, willow (*Salix viminalis*), poplar (*Populus monviso*), birch (*Betula pendula*), and oak (*Quercus robur*) grown on four TE-contaminated soils, a TE-spiked soil and an uncontaminated control soil. The aim of this study was to investigate the effect of trace elements on the growth and uptake of the aforementioned candidate species in a range of contaminated soils.

MATERIALS AND METHODS

Soils

The soils used in this study were taken from four polluted sites in Switzerland: Allmend, Losone, Dornach and Witzwil. Shooting ranges at Allmend, Luzern (Robinson *et al.*

Table 1 Physical and chemical properties six soils (mean ± standard variation, n = 5)

		Allmend	Dornach	Losone	Witzwil	WSL	Control
	sand	44	23	64	41	52	n.d.
	silt	34	39	29	28	36	n.d.
Texture (%)	clay	22	38	7	31	12	n.d.
$pH (CaCl_2)$		5.4 ± 0.2	7.2 ± 0.1	6.0 ± 0.1	6.77 ± 0.1	6.3 ± 0.1	6.2 ± 0.2
Corg (%)		2.1 ± 0.1	2.9 ± 0.1	4.3 ± 0.4	11.5 ± 0.7	1 ± 0.1	2.7 ± 0.2
$CaCO_3$ (%)		< 1	11.6 ± 0.5	1.3 ± 0.2	3.6 ± 0.4	3.7 ± 0.1	< 1
Electric conductivity ($\mu S\ cm^{-1}$)		80 ± 2	120 ± 4	134 ± 3	487 ± 3	174 ± 1	
C/N-ratio		16.7 ± 0.9	21.1 ± 1.5	24.3 ± 1.4	20 ± 0.9	36.3 ± 4.6	39.9 ± 6.6
Total Na ($g\ kg^{-1}$)		5.3 ± 0.9	2.42 ± 0.5	8.4 ± 0.9	2.1 ± 0.5	6.9 ± 0.7	0.6 ± 0.0
Total Mg ($g\ kg^{-1}$)		11.7 ± 0.3	7.4 ± 0.2	11.1 ± 0.3	6.0 ± 0.2	5.4 ± 0.2	1.7 ± 0.0
Total P ($g\ kg^{-1}$)		1.1 ± 0.0	1.6 ± 0.0	1.3 ± 0.0	2.8 ± 0.0	1.5 ± 0.1	1.5 ± 0.1
Total S ($g\ kg^{-1}$)		1.4 ± 0.1	0.7 ± 0.0	1.7 ± 0.1	3.8 ± 0.0	0.4 ± 0.0	3.4 ± 0.0
Total K ($g\ kg^{-1}$)		17.7 ± 0.2	15.1 ± 0.1	18.4 ± 0.2	10.4 ± 0.0	12.8 ± 0.1	5.3 ± 0.1
Total Ca ($g\ kg^{-1}$)		5.5 ± 0.0	49.6 ± 1.1	19.5 ± 0.8	39.2 ± 0.4	21.1 ± 1.8	19.9 ± 0.2
Total Fe ($g\ kg^{-1}$)		33.3 ± 0.6	33.9 ± 0.3	29.3 ± 0.6	41.4 ± 0.7	18.6 ± 0.5	3.7 ± 0.3
Total Mn ($mg\ kg^{-1}$)		701 ± 17	899 ± 9	464 ± 8	1146 ± 13	734 ± 17	379 ± 15
Total Ni ($mg\ kg^{-1}$)		41.4 ± 1.6	67.0 ± 2.5	61.3 ± 2.6	91.6 ± 1.7	26.1 ± 1.4	6.9 ± 0.1
Total Cu ($mg\ kg^{-1}$)		55.8 ± 4.7	653 ± 8	60.3 ± 3.3	750 ± 27	295 ± 6	39.2 ± 5.1
Total Zn ($mg\ kg^{-1}$)		153.8 ± 4.8	806 ± 9	99.4 ± 2.2	971 ± 13	1508 ± 48	79.9 ± 1.9
Total Cd ($mg\ kg^{-1}$)		0.5 ± 0.2	2.2 ± 0.2	0.4 ± 0.2	0.4 ± 0.0	6.4 ± 1.3	0.5 ± 0.0
Total Sb ($mg\ kg^{-1}$)		41.3 ± 13.6	1.7 ± 0.2	14.5 ± 1.5	36.1 ± 11.5	1.1 ± 0.2	0.0 ± 0.0
Total Pb ($mg\ kg^{-1}$)		2811 ± 263	66.6 ± 8.6	600 ± 53	878 ± 43	70.7 ± 1.9	16.8 ± 0.2

2008) and Losone (Knechtenhofer, Xifra, Scheinost *et al.* 2003) were heavily contaminated with Pb and Sb. The Allmend soil is a silty loam soil and with pH 5.4 the most acidic among the six soils and the highest Pb and Sb concentrations (Table 1). The Losone soil was a sandy loam soil with an organic carbon content of 4.3%. The Dornach soil was a clay loam, which had been heavily contaminated with Cu and Zn for about a century by particulate emissions from an adjacent brass smelter (Kayser *et al.* 2000). Besides Cu and Zn concentrations of approximately 650 $mg\ kg^{-1}$ and 800 $mg\ kg^{-1}$, respectively, the Dornach soil had high carbonate content (11.6%). The Witzwil soil was a clay loam from an agricultural field in Western Switzerland. It had been contaminated with Zn, Pb, and Cu by disposal of municipal wastes during the first half of the 20th century (Fässler *et al.* 2010), which also explains its high organic carbon content (11.5%). The WSL soil, a loam soil, had been artificially contaminated with dust from the brass smelter of Dornach, in a previous lysimeter experiment at WSL Birmensdorf (Switzerland) (Menon *et al.* 2005). The control soil was an uncontaminated soil substrate (technosol) consisting of peat bark humus and clay pot mixture, as it is regularly used for growing seedlings in the tree nursery of WSL (47°,21′,40 N; 8°,27′,19 S).

Soil Analysis

Soil samples were taken from the top 20 cm of the soil profiles at the respective sites of origin, dried at 40°C sieved to <2 mm and was then thoroughly mixed. Soil texture was determined using the hydrometer method after wet oxidation of the organic matter by means of hydrogen peroxide (FAL, RAC, and FAW 1996a). Organic matter content was determined using the dichromate method (FAL, RAC, and FAW 1996b). The carbonate content was measured by volumetric analysis of the CO_2 that evolved after addition of 4 M HCl to the soil. Total carbon (C_{tot}) and total nitrogen (N_{tot}) contents were determined by means of a CN-analyser (CNS-2000, Leco, USA). Electrical conductivity was determined according to DIN ISO 11265, and soil pH was measured in 0.01 M CaCl2 (FAL, RAC, and FAW, 1996d). Total soil TE concentrations were determined by means of X-ray fluorescence spectroscopy (Spectro X-lab 2000, Germany). Soluble soil TE were extracted with 0.1 M $NaNO_3$ (FAL, RAC, and FAW, 1996c). Filtrated extracts were analysed for TE by means of ICP-OES (Varian, Vista-MPX CCS simultaneous). For quality assurance, we analysed the two Wageningen soil standards, 989 and 951. Recoveries were 90% for Pb, Cd, Cu, and Zn.

Experimental Setup

Pot experiments were conducted in 2008 in the WSL tree nursery under ambient climate conditions at Birmensdorf between May and September. Ten L plastic pots with six small holes at the bottom, were filled with approximately 9 kg air-dried and 1 cm sieved soil. A tray was placed under each pot. The soil was fertilized using Osmocote 6 M at rates recommended by the manufacturer, resulting in the addition of 500 mg N, 100 mg P, and 200 mg K kg^{-1} of soil. Pots were irrigated 3–4 times per week.

Each soil and tree species, was replicated six times for a total of 144 pots. The willows and poplars were planted as approximately 30 cm long unrooted cuttings weighing of 9.5 ± 1.5 g and 20.1 ± 2.5 g, respectively. The birches were planted as approximately 20 cm tall 1.5-year-old seedlings grown from seeds, and the oaks were planted as approximately 35 cm tall 3-year-old seedlings grown from acorns. The shoot and root dry weights were 0.32 ± 0.1 g and 0.43 ± 0.1 g, respectively, for the birches and 3.0 ± 1.0 g and 2.26 ± 1.0 g, respectively, for the oaks at planting. The 144 pots were set up on a grid in a completely randomised design.

Plant Harvest, Biomass, and Analysis

All trees were harvested after 4 months of growth. The aboveground biomass was sampled by cutting stems at 5 cm above ground and then divided into leaves and stems (including branches). Roots were cleared from the substrate by washing. Poplar and willow roots were separated from the cutting. All plant samples were subsequently washed with tap and deionised water and then dried at 60°C, until constant weight was obtained. After weighing, the dry samples were ground using a Retsch ZM-200 centrifugal titanium mill. Foliage, stem and roots samples were analysed for TE by means of X-ray fluorescence spectroscopy (Spectro X-lab 2000, Germany). For quality assurance, we analyzed certified plant reference material (Virginia tobacco leaves CTA-VTL-2, Polish Reference Material). Recoveries were >90% for Pb, Cu, Cd, Fe, and Zn; and 90–110% for Ca, Mg, Mn, K, and P. For Sb, approximately 0.2 g subsample of each ground plant sample was digested with 2 mL HNO_3 (65%) and 4 mL H_2O_2 (30%) using a microwave digester (lavis ETHOS *MLS* GmbH, Leutkirch, Germany) to prevent Sb volatilization. The digests were diluted to 25 mL with nanopur water. The extracts were analysed for Sb by ICP-MS (Variam, 810-MS) with a detection limit of 0.0002 mg L^{-1}. For quality assurance, we analysed certified plant reference material (Virginia tobacco leaves CTA-VTL-2, Polish Reference 147 Material). Recoveries Sb were >90%.

Statistical Analysis

All statistical analyses were carried out by means of SPSS. Treatment effects were determined by analysis of variance (ANOVA). Differences were considered significant if $p < 0.05$.

RESULTS

Trace Elements Availability

The five contaminated soils varied considerably in total metal concentrations, which ranged for Cu between 55.8 and 750 mg kg^{-1}, for Zn between 99.4 and 1508 mg kg^{-1}, for Pb between 66.9 and 2811 mg kg^{-1} and for Cd between 0.5 and 6.4 mg kg^{-1} as well as in their physical and chemical properties (Table 1). The physical and chemical properties of the soil also had an effect on the $NaNO_3$-available TE concentration in these soils (Table 2). Of all the soils, Allmend was had the lowest pH of 5.4 and highest $NaNO_3$-available fraction (percentage of available metal concentration/total metal concentration) of Mn (0.024%), Ca (40%) and Pb (1.5%). Dornach's high pH of 7.2 caused by its high $CaCO_3$-content of 11.6% and high clay content of 38% resulted in low Cu (0.12%), Zn (0.014%), Pb (below detection, b.d.) and Mn (0.003%) available fractions. With an organic carbon content of 11.5%, the Witzwil soil contained 2.6 to 11.5-fold higher organic carbon than the other soils and also had the lowest $NaNO_3$-available fraction of Cu (0.07%), Zn (0.014%), and Pb (b.d.). Losone had the highest Sb available fraction (0.46%), followed by Witzwil and Allmend with 0.15% and 0.14%, respectively.

Plant Dry Weight

On the contaminated soils willow, poplar and birch growth was much reduced (Figure 1) with respect to the control. In addition we found foliar necrosis and chlorosis on the Dornach and WSL soils in all three species, however the extent of the reactions varied among the tree species. In contrast, neither a growth effect nor toxicity symptoms was observed in oak. For the other species, the strongest growth inhibition was on the WSL soil, with average biomass of 39.1 g, 3.2 g, and 25.7 g per tree in poplar, willow and birch, respectively. Dry weight of poplar, willow and birch growing on Dornach soil was significantly reduced by 1.7-, 6.3-, and 3-fold, respectively.

The roots were similarly adversely affected by the contaminated soils. Contrary to shoot biomass, root biomass on the control soil varied significantly among the three tree species, with 78.5, 25.9 and 51.1 g for willow, poplar and birch, respectively. In willow, poplar, and birch, shoot, and root growth was more severely inhibited on the WSL soil. Also, the biomass of willow and poplar cuttings was affected by all treatment soils. Compared to the initial weight of the planted cuttings, only the willow cuttings grown on the control (33.7 g), Losone (21.4 g), and Witzwil (27.7 g) soil and the poplar cuttings grown on the control (42.2 g) and Dornach (29.5 g) soil significantly increased their dry weight (Figure 1).

Metal Accumulation and Allocation

Tree species patterns. Cadmium, Zn, Pb, Cu, Sb, Mn, Ca, Fe, Mg, Na, and K accumulations in foliage and wood varied considerably between tree species (Table 3). With the exception of Pb, TE accumulation in foliage was higher than in wood, especially

Table 2 $NaNO_3$-available metal concentrations (mg kg^{-1}) of six soils (mean ± standard variation, n = 5)

Soil	Cu	Cd	Zn	Pb	Sb	Mn	Fe	Ca	Mg	K
Allmend	0.03 ± 0.0	0.02 ± 0.0	0.15 ± 0.00	1.6 ± 0.06	0.06 ± 0.00	1.2 ± 0.04	0.07 ± 0.01	1990 ± 25	38.8 ± 0.25	16.9 ± 0.95
Dornach	0.81 ± 0.08	0.03 ± 0.0	0.12 ± 0.03	b.d.	b.d.	0.03 ± 0.03	0.06 ± 0.03	2790 ± 55	23.6 ± 0.81	37.8 ± 0.98
Losone	0.02 ± 0.0	b.d.	0.05 ± 0.02	b.d.	0.07 ± 0.00	0.3 ± 0.02	0.14 ± 0.01	1280 ± 70	48.3 ± 2.6	72.5 ± 3.2
Witzwil	0.53 ± 0.02	b.d.	0.13 ± 0.01	b.d.	0.06 ± 0.00	0.1 ± 0.02	0.30 ± 0.03	4370 ± 320	57.5 ± 2.0	141.1 ± 1.7
WSL	0.37 ± 0.02	0.10 ± 0.02	5.9 ± 0.13	b.d.	b.d.	0.08 ± 0.0	0.5 ± 0.02	1060 ± 350	23.28 ± 0.86	43.1 ± 0.83
Control	0.01 ± 0.0	b.d.	0.1 ± 0.02	b.d.	b.d.	0.5 ± 0.08	0.21 ± 0.05	1350 ± 120	58.5 ± 5.4	35.6 ± 5.3

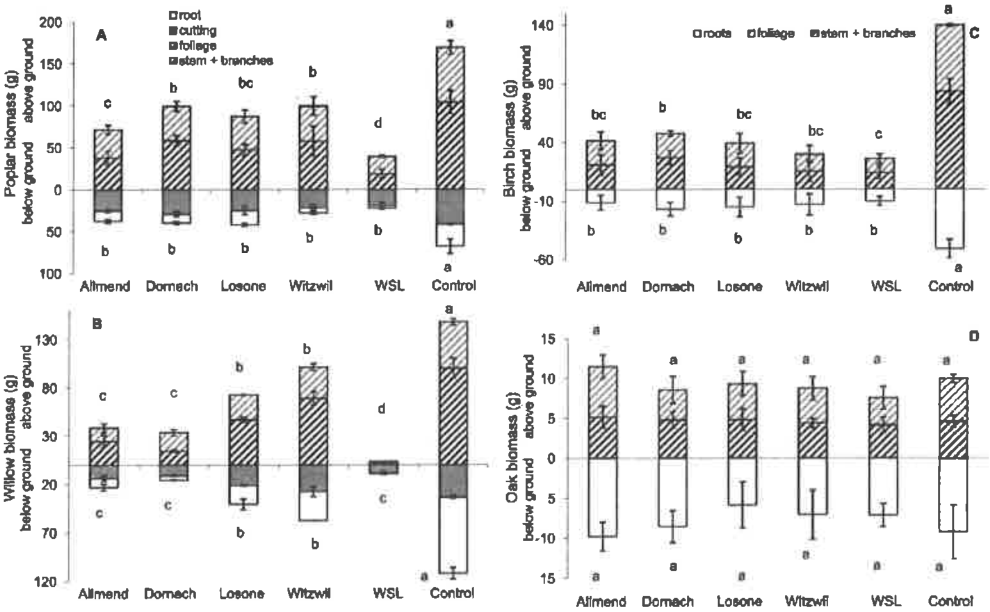

Figure 1 Shoot and root dry biomass production (mean of standard variation, n = 6) of poplar (A), willow (B), birch (C), and oak (D) after four months of growth, on the soils of Allmend, Dornach, Losone, Witzwil, WSL, and control. (Different letters show significant difference.)

at high TE soil concentrations. Although the total and available soil element concentrations were significantly different in the five contaminated soils (Tables 1 and 2), distinctive patterns could be observed concerning the element accumulation by the four investigated tree species.

Cadmium accumulation decreased in the following order: willow > poplar > birch > oak, which agrees with Hermle *et al.* (2006) where willow accumulated more Cd than birch. Willow had the highest Cd and Zn concentrations in foliage and wood on all contaminated soils, with ranges of 3.4–40.9 mg kg^{-1} Cd and 286–2088 mg kg^{-1} Zn in foliage and ranges of 3.4 to 40.9 mg kg^{-1} for Cd and 88.5 to 414 mg kg^{-1} for Zn in wood (Table 3). Cadmium concentrations in wood and foliage of willow agreed with those of Ledin (1998), Eriksson and Ledin (1999), and Hermle *et al.* (2006). Foliage and wood Sb concentrations were very low, ranging from 0.07 to 0.38 and 0.02 to 0.10 mg kg^{-1}, respectively, agreeing with the concentrations found by Robinson *et al.* (2008). Willow and poplar foliage had significantly higher Ca and K foliage concentrations than birch and oak, with concentrations ranging between 12511–44431 mg kg^{-1} and 12625–37325 mg kg^{-1}, respectively. However in wood, poplar had even higher K concentrations than willow, with concentrations ranging from 6721 to 10770 mg kg^{-1} (Table 3). These values are comparable with the bark and wood Ca and K concentrations stated by Tharakan *et al.* (2003).

On the two shooting range soils contaminated with high Pb concentrations, birch had the highest wood (135 mg kg^{-1}) and foliar (78 mg kg^{-1}) Pb concentrations, and birch also had the highest Fe concentrations in foliage on all contaminated soils. Oak had the highest Mn, Fe, Ca and Mg wood concentrations on all soils but WSL. On the other hand, oak had the lowest Cd and Zn concentrations, which for Cd were below or close to the detection limit (<0.7 mg kg^{-1}), in foliage and wood.

Table 3 Foliage and wood metal concentrations of willow, poplar, birch and oak on six soils (mean ± standard variation, n = 3)

Soil	Tree species	Foliage metal concentrations ($mg\ kg^{-1}$)										
		Cu	Cd	Zn	Pb	Mn	Fe	Sb	Ca	Mg	K	Na
Allmend	willow	14.4 ± 1.4	6.3 ± 0.9	616 ± 112	53.2 ± 10.5	162 ± 12.9	73.4 ± 7.4	0.13 ± 0.01	14800 ± 2130	1510 ± 280	12600 ± 1100	362 ± 46.1
	poplar	15.2 ± 1.7	2.1 ± 0.54	224 ± 50.6	31.7 ± 10.1	117 ± 11.1	54.4 ± 4.5	0.07 ± 0.01	12500 ± 1040	1650 ± 160	17700 ± 2000	157 ± 53
	birch	14.9 ± 2.0	1.0 ± 0.18	260 ± 45.5	78.3 ± 16.4	326 ± 65.1	114 ± 21.5	0.09 ± 0.04	7000 ± 900	1630 ± 180	12000 ± 1700	186 ± 84.3
	oak	19.9 ± 4.8	b.d.	43.4 ± 7.8	40.2 ± 6.7	197 ± 48.9	56.4 ± 5.2	0.08 ± 0.01	10000 ± 1500	1740 ± 880	7510 ± 1100	199 ± 90.3
Dornach	willow	34.9 ± 2.0	14.7 ± 2.7	1150 ± 178	b.d.	122 ± 15.1	71.7 ± 20.6	b.d.	44400 ± 5200	2600 ± 160	37300 ± 3600	587 ± 63.7
	poplar	14.6 ± 3.1	5.9 ± 0.63	361 ± 36.2	b.d.	72.0 ± 13.3	43.7 ± 3.9	b.d	18300 ± 1000	1510 ± 130	18500 ± 1900	355 ± 114
	birch	19.5 ± 5.3	b.d.	348 ± 76.7	b.d.	74.5 ± 18.3	93.5 ± 12.1	b.d	11200 ± 1800	2100 ± 396	11700 ± 1520	181 ± 39.9
	oak	28.9 ± 5.3	b.d.	47.6 ± 15.3	b.d.	126 ± 144	43.4 ± 8.6	b.d	28200 ± 4500	2160 ± 1580	9100 ± 1300	203 ± 72.5
Losone	willow	11.7 ± 1.1	3.35 ± 0.56	286 ± 24.1	3.0 ± 0.74	149 ± 21.4	89.3 ± 4.8	0.38 ± 0.01	14000 ± 910	1320 ± 175	18400 ± 840	288 ± 80.4
	poplar	13.7 ± 1.6	1.6 ± 0.37	91.8 ± 11.5	1.9 ± 0.63	77.9 ± 7.7	50.9 ± 3.4	0.19 ± 0.03	14300 ± 910	1650 ± 150	22800 ± 1240	243 ± 83.4
	birch	14.4 ± 2.7	b.d.	135 ± 27.9	5.2 ± 1.6	87.1 ± 11.1	111 ± 12.9	0.32 ± 0.02	7930 ± 1040	2000 ± 175	11600 ± 2120	119 ± 11.3
	oak	27.4 ± 6.9	b.d.	27.4 ± 6.9	4.9 ± 1.3	95.8 ± 42.0	52.4 ± 14.3	0.24 ± 0.05	8220 ± 1700	1960 ± 400	8000 ± 1270	185 ± 84.9
Witzwil	willow	11.9 ± 3.5	3.78 ± 1.15	555 ± 147	b.d.	150 ± 24.6	79.8 ± 12.0	0.18 ± 0.01	17700 ± 2300	1120 ± 195	18400 ± 2800	406 ± 67.7
	poplar	9.0 ± 2.0	2.1 ± 0.32	249 ± 48.2	b.d.	82.1 ± 13.1	48.6 ± 5.0	0.08 ± 0.01	14200 ± 2020	1150 ± 150	22500 ± 3110	277 ± 157
	birch	16.3 ± 5.8	0.73 ± 0.34	305 ± 147	2.7 ± 0.5	128 ± 53.9	139 ± 49.9	0.19 ± 0.01	9160 ± 2400	1600 ± 320	11000 ± 4140	190 ± 31.4
	oak	54.0 ± 17.5	b.d.	54.1 ± 17.5	1.7 ± 0.3	99.4 ± 24.5	60.5 ± 25.6	0.10 ± 0.03	16400 ± 4520	2500 ± 690	9250 ± 780	173 ± 31.3
WSL	willow	25.5 ± 17.6	40.9 ± 12.3	2080 ± 110	2.2 ± 1.3	67.8 ± 49.8	121 ± 33	b.d	23800 ± 21000	1800 ± 1000	19900 ± 20300	1120 ± 360
	poplar	19.1 ± 11.3	29.9 ± 6.1	1690 ± 349	2.7 ± 1.5	82.9 ± 6.8	54.1 ± 29.4	b.d	24800 ± 5260	2110 ± 250	31500 ± 4860	757 ± 141
	birch	16.4 ± 3.1	14.3 ± 3.2	1040 ± 135	1.6 ± 1.0	106 ± 19.5	164 ± 62.6	b.d	10000 ± 1830	1930 ± 240	12300 ± 805	332 ± 64.2
	oak	188 ± 78.9	0.6 ± 0.05	188 ± 78.9	1.1 ± 0.5	102 ± 28.9	90.4 ± 8.3	b.d	21100 ± 10600	3140 ± 1250	10300 ± 1600	299 ± 78.9
Control	willow	11.2 ± 2.6	0.62 ± 0.15	136 ± 22.8	b.d.	150 ± 13.8	76.9 ± 12.1	b.d	11500 ± 1000	1650 ± 250	20200 ± 1260	240 ± 61.7
	poplar	4.3 ± 0.87	0.6 ± 0.06	147 ± 17.2	b.d.	129 ± 21.1	48.9 ± 7.1	b.d	13500 ± 3500	1920 ± 150	21400 ± 1130	280 ± 46.6
	birch	11.5 ± 2.4	1.0 ± 0.16	248 ± 58.3	b.d.	310 ± 55.4	98.6 ± 12.6	b.d	7500 ± 980	1600 ± 270	15000 ± 297	140 ± 30.3
	oak	12.2 ± 9.1	b.d.	42.8 ± 8.4	b.d.	255 ± 100	64.4 ± 16.4	b.d	7320 ± 1030	1840 ± 430	10100 ± 1930	150 ± 53.4

Soil	Tree species	Wood metal concentrations (mg kg^{-1})										
		Cu	Cd	Zn	Pb	Mn	Fe	Sb	Ca	Mg	K	Na
Allmend	willow	7.5 ± 0.85	3.2 ± 0.95	141 ± 19.3	76.4 ± 7.6	27.4 ± 3.6	9.8 ± 1.9	0.02 ± 0.01	5370 ± 467	502 ± 32.0	3600 ± 418	137 ± 27.4
	poplar	9.7 ± 1.7	1.2 ± 0.39	80.1 ± 16.9	56.1 ± 13.9	28.9 ± 3.9	8.3 ± 1.7	0.03 ± 0.01	4650 ± 557	756 ± 140	6720 ± 1919	171 ± 39.1
	birch	7.0 ± 0.54	0.95 ± 0.35	133 ± 13.1	134 ± 25.6	77.8 ± 20.3	18.2 ± 6.3	0.04 ± 0.01	3900 ± 414	587 ± 31.3	5080 ± 705	133 ± 66.0
	oak	10.2 ± 2.1	b.d.	55.5 ± 12.9	38.9 ± 5.5	128 ± 24.3	20.9 ± 3.5	0.08 ± 0.01	10400 ± 1200	1150 ± 317	4790 ± 615	136 ± 33.8
Dornach	willow	21.6 ± 2.3	4.1 ± 0.50	150 ± 30.5	2.5 ± 0.77	15.7 ± 1.1	8.2 ± 3.2	b.d.	7670 ± 622	495 ± 74.2	8010 ± 940	245 ± 89.9
	poplar	6.6 ± 0.88	2.0 ± 0.03	71.3 ± 4.6	b.d.	15.1 ± 1.2	5.4 ± 1.4	b.d	5440 ± 663	609 ± 69.9	7750 ± 661	182 ± 36.9
	birch	7.4 ± 1.7	0.68 ± 0.10	131 ± 19.2	b.d.	17.5 ± 1.8	10.7 ± 1.8	b.d	4750 ± 1010	539 ± 23.2	4260 ± 717	149 ± 34.8
	oak	13.2 ± 4.0	b.d.	53.6 ± 15.0	b.d.	31.0 ± 4.0	22.5 ± 8.2	b.d	14100 ± 2310	1170 ± 78.4	4390 ± 532	135 ± 10.7
Losone	willow	6.6 ± 0.70	1.9 ± 0.12	88.5 ± 5.9	5.3 ± 1.2	26.8 ± 3.9	9.5 ± 1.8	0.08 ± 0.01	5090 ± 352	512 ± 55.9	5400 ± 545	162 ± 12.7
	poplar	7.2 ± 0.74	0.95 ± 0.12	34.3 ± 2.5	2.55 ± 0.79	17.8 ± 1.2	6.1 ± 0.84	0.06 ± 0.01	2170 ± 576	623 ± 60.4	8780 ± 1450	111 ± 19.3
	birch	6.5 ± 1.2	b.d.	85.9 ± 11.5	13.1 ± 4.8	24.2 ± 3.5	12.2 ± 2.2	0.08 ± 0.01	4000 ± 640	537 ± 53.4	5400 ± 742	146 ± 43.8
	oak	9.2 ± 0.81	b.d.	30.8 ± 6.8	5.6 ± 1.9	50.5 ± 16.8	27.8 ± 14.5	0.10 ± 0.02	8370 ± 1780	1080 ± 81.9	4560 ± 668	125 ± 22.4
Witzwil	willow	7.7 ± 1.4	1.8 ± 0.33	132 ± 27.7	b.d.	32.7 ± 8.7	8.9 ± 3.0	0.04 ± 0.01	5740 ± 547	553 ± 69.8	5109 ± 1203	160 ± 66.6
	poplar	4.7 ± 1.0	0.98 ± 0.06	54.7 ± 14.2	b.d.	19.0 ± 1.6	6.1 ± 1.2	0.05 ± 0.01	4765 ± 757	552 ± 85.6	9034 ± 2946	125.2 ± 32.5
	birch	8.7 ± 3.6	0.72 ± 0.18	142 ± 34.9	1.5 ± 0.68	34.5 ± 18.2	22.1 ± 11.9	0.06 ± 0.01	5090 ± 1170	501 ± 109	5249 ± 4151	147 ± 41.3
	oak	10.5 ± 3.4	b.d.	63.5 ± 17.1	1.52 ± 0.52	47.8 ± 10.1	37.4 ± 11.4	0.07 ± 0.02	11200 ± 1900	1280 ± 115	5996 ± 484	105 ± 8.3
WSL	willow	13.4 ± 3.9	13.8 ± 3.9	414 ± 132	1.9 ± 0.18	24.4 ± 8.2	40.1 ± 24.1	b.d	9370 ± 1690	624 ± 153	6260 ± 3536	1143 ± 263
	poplar	9.4 ± 2.1	15.4 ± 6.5	481 ± 129	b.d.	19.3 ± 3.6	15.7 ± 11.4	b.d	13700 ± 4300	1040 ± 244	10800 ± 3200	415 ± 103
	birch	5.7 ± 1.9	8.7 ± 1.6	351 ± 78.9	1.0 ± 0.52	24.1 ± 7.8	13.8 ± 5.9	b.d	4050 ± 1520	438 ± 123	4150 ± 903	204 ± 33.9
	oak	14.1 ± 2.6	0.72 ± 0.18	138 ± 95.4	1.8 ± 0.43	30.2 ± 11.3	21.0 ± 7.5	b.d	9850 ± 5600	1050 ± 651	3640 ± 2115	178 ± 200
Control	willow	2.7 ± 0.60	0.8 ± 0.53	89.3 ± 8.8	b.d.	32.8 ± 5.1	5.4 ± 0.70	b.d	3960 ± 490	539 ± 60.1	6050 ± 482	149 ± 77.3
	poplar	1.1 ± 0.03	b.d.	59.5 ± 6.7	b.d.	26.1 ± 2.7	6.4 ± 1.3	b.d	5020 ± 569	667 ± 98.1	7900 ± 1129	196 ± 5.3
	birch	2.5 ± 0.60	b.d.	132 ± 18.5	b.d.	40.2 ± 4.5	16.9 ± 6.7	b.d	2980 ± 369	482 ± 28.8	5400 ± 432	132 ± 0.71
	oak	5.7 ± 1.9	b.d.	71.2 ± 22.7	b.d.	173 ± 63.8	56.3 ± 16.3	b.d	9050 ± 1510	1300 ± 242	7600 ± 1746	165 ± 45.3

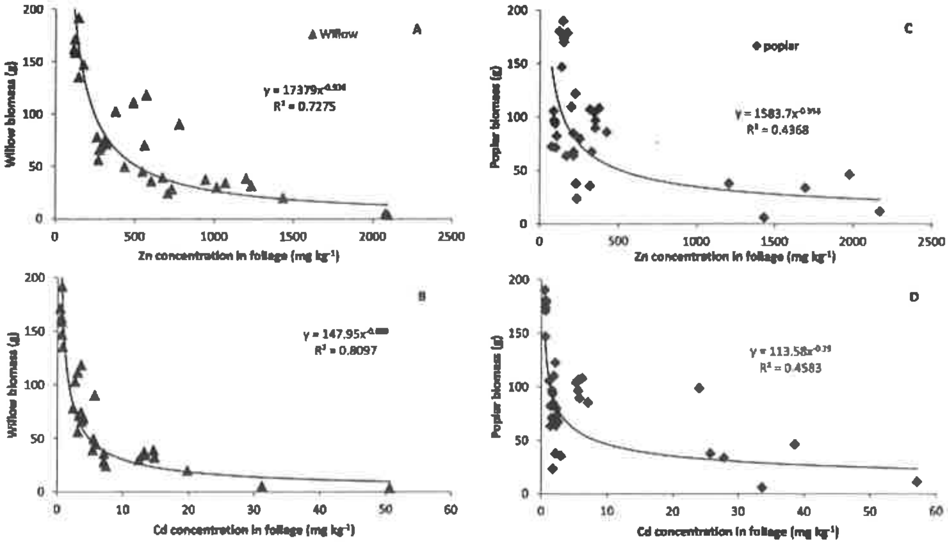

Figure 2 Effect of foliage Zn and Cd concentration, in willow (A, B) and poplar (C, D), on whole-trees dry biomass production (n = 6).

Metal allocation. For stem and foliage with Cd concentrations exceeding 1 mg kg^{-1} Cd concentrations in foliage were 2–3 times higher than in the stems, which agrees with Hermle *et al.* (2006) and Riddell-Black (1994) who found foliage/stem Cd concentration quotients of 3–5. Potassium showed similar allocation patterns as Zn and Cd. Potassium concentrations in willow, poplar and birch foliage were 2–4 fold higher than in stems. This is in agreement with Giachetti and Sebastiani (2006) who, although found lower concentrations than here, showed the same concentrations ratio between leaves and stem. In contrast to the other investigated TE, Pb accumulation was higher in the stem than in the foliage (Table 3), as noted by Hasselgren (1999), and in contradiction to Jensen *et al.* (2009) who found higher Pb concentrations in willow leaves than in the stems and twigs.

DISCUSSION

Under non-metal stress conditions willow and poplar would be favoured for phytostabilization/phytomanagement due to their high aboveground biomass production. However, a general statement for various sites is difficult to make, as the biomass production depends on the site characteristics. Besides nutrient supply, biomass production of various tree species responds differently to the TE concentrations in the soil and their uptake into the plant (Figure 1). In this study willow and poplar biomass reduction for was correlated with Cd ($R^2 = 0.81$ and 0.46) and Zn ($R^2 = 0.72$ and 0.44) leaf concentrations (Figure 2). Of these two TE, Cd had the highest impact. According to Kabata-Pendias and Pendias (2001) Cd is approximately 17-fold more toxic than Zn. Although Pb, Cu, and Ni are 3 to 10-fold more toxic to plant organisms than Zn, their concentrations in willow and poplar leaves were too low (Table 3) to affect growth. For oak and birch, biomass was not correlated with any TE.

Although oak was the most tolerant species and could thus be planted on soils with very high TE soil concentrations, it had a very low root biomass production (Figure 1). An extended root system is favourable for preventing soil erosion and subsequent contaminant dispersion. The net growth of root mass of willow, poplar and birch was between 2- and 15-fold higher than that of oak during the experiment. Thus, oak would be less suitable than willow, poplar, and birch for reducing soil erosion.

Trace element accumulation in foliage and wood is crucial in the choice of tree species for combined phytostabilization/phytomanagement. Besides wind and water erosion and dispersion of foliage (Perronnet *et al.* 2000), risks could arise from the consumption of plant parts by animals, as TE could thus enter the food chain. According to Swiss regulations (FOEN 2005), the maximum allowed TE concentration in fodder (dry weight) are: 150 mg kg^{-1} Zn, 40 mg kg^{-1} Pb, 15–35 mg kg^{-1} Cu, and 1 mg kg^{-1} Cd. Copper concentrations in the foliage and stem of the four tree species never exceeded these values. Foliage Zn concentrations surpassed the Zn tolerance value on all contaminated soils for willow, poplar and birch, even on the control soil. The birch foliage surpassed the guideline concentrations only for Zn, thus posing a lower risk for herbivores due to its low Cd concentrations. Oak leaves remained, with the exception of the WSL soil, below the Zn tolerance values, and because of the unpalatable tannins present in high concentrations, are not eaten by deer and other animals. Tannins affect the availability of nutrients and even produce toxic effects when animals consume high amounts of oak leaves (Makkar, Singh, and Dawra 1988). To protect the plantation itself from herbivores and in order to protect the herbivores from the TE in the foliage, in the first year of the plantation a protective fence would be an option. Lead foliage and stem concentrations surpassed the guideline levels only for the trees growing on the Allmend shooting range soil. Thus, when the criteria shoot and root biomass as well as TE accumulation are taken into consideration, willows and poplars would be the first choice on soils with low and moderate Zn and Cd soil concentrations. On soils with elevated Zn and Cd concentration, birch would be the most suitable tree species, but it would be unsuitable for soils with high soil concentrations of Pb because of its high accumulation of Pb.

Entry of foliage and woody parts into the food chain would be of concern only in the first years of growth. Once the foliage is out of reach to herbivores risks of food chain transfer would be minimized. In that case the intake of metals will probably increase due to the intake of soil thus resulting in a probably higher risk than that posed by the tree foliage (Smith, Abrahams, Dagleish *et al.* 2009). Entry of TE into the food chain can also occur through invertebrates, which were reported to bioaccumulate Pb on shooting ranges (Migliorini *et al.* 2004). Only in winter when foliage falls from the trees could a risk occur. However, willow, poplar and birch foliage which surpassed only the Zn tolerance values could be collected and used as fodder additives, to alleviate animal Zn deficiency.

Metal concentrations in wood are important when the wood is used commercially. If the wood is gasified for energy production, high concentrations of inorganic elements, such as Ca, K, Mg and Na can promote the formation of gaseous species and char at the expense of bio-oil yield (Patwardhan *et al.* 2010). According to Obernberger *et al.* (2006), typical wood fuels from short rotation coppice willow (from uncontaminated areas) contain 3000 mg kg^{-1} K and 5000 mg kg^{-1} Ca. In this experiment, some trees species were occasionally below 5000 mg kg^{-1} Ca, but none of the tree species were below 3000 mg kg^{-1} K. However, our results are uptakes by four months old trees, which have a high bark to wood ratio. Metal concentrations are significantly higher in bark than in wood (Obernberger *et al.* 2006; Evangelou, *et al.* 2012), thus a favourable reduction of the bark to wood ratio is likely in older trees. Therefore, Adler, Dimitriou, Aronsson *et al.* (2008), proposed an increase in the age of coppiced trees from 1–2 y to 3 y. For example harvestable shoot biomass of willows grown as a few large stems would have a better wood fuel quality, compared to harvestable shoot biomass of many small stems. Thus when biomass is intended for biofuel/bioenergy production, an increased length of cutting cycle would improve the wood fuel quality. The wood could also be used for furniture, construction or industrial purposes. According to Swiss legislation and EPF industry standard wood, intended for the market should not exceed concentrations of 50 mg kg^{-1} Cd, 90 mg kg^{-1} Pb and 40 mg kg^{-1} Cu (EPF; Chem RRV 2005). Since birch grown on Allmend soil exceeded one of these values, the biomass deriving from these contaminated soils could also be used for industrial or furniture products.

CONCLUSION

The four tree species showed distinctly different biomass yield responses to the contaminated soil. However, they had specific TE uptakes independent of the soils. Willow had the highest Zn and Cd uptakes and birch had the highest Pb uptake, on sites with high soil concentrations of these elements. Our results indicate that tree selection should be site specific. Birch is most suitable for biomass production combined with phytostabilization for soils with high Cd and Zn but low Pb concentrations, while poplars and willows could stabilise soils with high Cu and Pb but not very high Zn and Cd concentrations. Birch, however, cannot be coppiced and is not feasible for short rotations. Oak was most tolerant to the TE contamination, but due to its low root and shoot biomass production, is unsuitable for phytomanagement purposes. However, if connected with an economic output, other than biofuels, oak could be preferable as its wood is valuable. Independent of the tree species used to combine phytostabilization and production of biomass for bioenergy safe, a constant monitoring will be necessary to control the risk of TE distribution though soil erosion and entrance of TE to the food chain.

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