Trace element accumulation by poplars and willows used for stock fodder

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Abstract Poplars and willows can accumulate high concentrations of some trace elements. We investigated the extent and nature of B, Cd, Co, Cu, Fe, Mn, and Zn accumulation in some commercial poplar and willow clones by using a lysimeter experiment and field collection of data. Trace element accumulation was a function of leaf age and the variety or species of tree. Leaf B, Cd, Mn, and Zn concentrations increased throughout the season, while Cu decreased and Fe remained unchanged. Poplars and willows accumulated high concentrations of the trace elements tested, relative to pasture. The accumulation of Cd is of concern, especially in willows. Stock exposure to Cd can be managed by judicious clone selection, harvesting young shoots, or harvesting early in the season. Poplars and willows may be used as feed supplements to increase Co and Zn intake by livestock. The varieties 'Yeogi' and 'Crow's Nest' accumulated the highest concentrations of Co, yet their Cd concentrations were not significantly higher than pasture.

Keywords heavy metal; nutrition; toxicity; clonal variation; cadmium

INTRODUCTION

As certain areas in New Zealand become consistently drier under a prevailing El Niño weather pattern (Plummer et al. 1999), deep-rooting tree species that can avoid periods of drought will take on increasing importance as stock fodder. Poplars and willows are used extensively in New Zealand for soil conservation and supplementary stock fodder during times of drought (Wilkinson et al. 1999). Both foliage and small twigs can be browsed by sheep and cattle (Hathaway 1986; Douglas et al. 1996).

In addition to providing an emergency food source, poplars and willows as stock feed have proven health benefits including an improvement in fecundity (Barry & Kemp 2001). These benefits may be derived from high protein, tannin or trace element concentrations.

In some areas, New Zealand pasture lands are deficient in essential trace elements, including Co, Zn, and Cu (Lee et al. 1999). Poplars and willows have been shown to accumulate large amounts of Zn relative to pasture species (Hinchman et al. 1996). The possibility exists, therefore, that poplars and willows could be used to provide trace element supplements in areas where the soils are deficient.

Trees used for stock fodder may also accumulate a range of toxic elements. Feeding such fodder to livestock may elevate levels of contaminants, such as Cd, in meat products. This could have deleterious effects on humans and may be used as a non-tariff trade barrier.

Most New Zealand pasture lands have elevated Cd concentrations due to repeated applications of Cd-rich superphosphate fertiliser (Bramley 1990).

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The average concentration of Cd in dry topsoil of New Zealand pasture lands is 0.44 μ g g⁻¹ (Roberts et al. 1994).

Robinson et al. (2000) showed that the commonly used New Zealand varieties of poplar, 'Kawa' (*Populus deltoides* × *P. yunnanensis*), and willow, 'Tangoio' (*Salix matsudana* × *S. alba*), accumulated Cd at levels of up to 14 μ g g⁻¹ in the dry leaves when grown in a soil containing just 0.6 μ g g⁻¹ of this element. This concentration is above levels (1–5 μ g g⁻¹) that adversely affect livestock (Underwood & Suttle 1999). Thus, Cd accumulation by poplar and willows that might be used for fodder is of concern.

Riddell-Black et al. (1997) and Laureysens et al. (2004) demonstrated that there is a wide variation in the ability of different willow clones to accumulate heavy metals in bark and wood. By using hydroponic experiments, Punshon & Dickinson (1999) showed interclonal variation in tolerance to Cu, Cd, Ni, and Zn, as well as in the uptake of Cu. Granel et al. (2002) demonstrated large genetic differences in the uptake of trace elements by willow clones. In the case of Cd, there was a 12-fold difference between the lowest and highest leaf Cd concentrations for sibling plants growing in identical environments.

Inter-clone differences in trace element accumulation could be exploited by breeding clones that have a high uptake of essential elements such as Co and Zn, and minimal uptake of any toxic heavy metals, such as Cd, that may be present. The trees may also be managed so that leaf material is harvested at a time when it has the best trace element balance for stock. Laureysens et al. (2004) showed that for some elements, there were significant differences between young, mature, and senescing leaves.

Concentration in drainage water

Average amount (g) lost through

leaching per lysimeter

 $(\mu g m l^{-1})$

The aim of this study was to determine the extent and nature of the uptake of B, Cd, Co, Cu, Fe, and Mn by commonly used New Zealand poplar and willow clones.

MATERIALS AND METHODS

Greenhouse experiment

A greenhouse experiment was set up in order to determine the accumulation of trace elements as a function of time. Sawdust was used *inter alia* in the lysimeters because it provides a homogeneous plant-growth medium and provides near-constant level of available trace elements over the time of the experiment.

Seven thousand litres of sawdust were obtained from a wood-waste pile and homogenised using a digger. The bulk density of the sawdust was 0.23, with a water holding capacity of 420 ml kg⁻¹. The total and soluble trace element concentrations in the sawdust are given in Table 1. In October 2000, eight 800 litre weighing lysimeters were filled with the sawdust, and a 1 m long poplar pole of the clone 'Toa' (Populus euramericana × P. yunnanensis) planted in each. The lysimeters were fertilised with Hoagland's solution (Hoagland & Arnon 1950) at three concentrations: low (2 lysimeters), medium (3 lysimeters), and high (3 lysimeters). Fertilisation occurred from March to September. The total amounts of N, P, and K applied to each lysimeter were (in grams): low 10 N, 1.5 P, 11 K, medium 20 N, 3 P, 22 K, and high 30 N, 4.5 P, 33 K. Table 1 shows the amounts of B, Cu, Fe, Mn, and Zn that were added

0 1	0 0	0					
Element	В	Cd	Co	Cu	Fe	Mn	Zn
Total concentration ($\mu g g^{-1}$)	44.5	8.1	31.9	140	4620	144	63.1
Average amount per lysimeter (g)	7.12	1.3	5.1	22.4	739	23	10.1
Total added (low, medium, high) in	0.024	_	_	0.001	0.024	0.024	0.002
Hoaglands' solution (g)	0.048			0.002	0.048	0.048	0.005
8	0.071			0.003	0.071	0.071	0.007

0.21

(0.01)

0.05

< 0.01

0.43

(0.07)

0.1

17.8

(1.0)

4.24

1.3

(0.1)

0.31

0.9

(0.2)

0.21

2.47

(0.09)

0.59

Table 1 Initial trace element concentrations in the sawdust ($\mu g g^{-1} dry wt$) and the average concentrations ($\mu g ml^{-1}$)in drainage water samples collected during the growing season. Values in brackets are the SEM.

in the Hoagland's solution. In all cases, the amount of trace element added was less than 1% of the total trace element content of the lysimeter. These plant nutrient treatments were used to determine effect of substrate fertility on the foliar trace element concentrations.

The lysimeters were given a measured amount of water daily to maintain their volumetric water content between 0.50 and 0.60. Drainage was collected weekly, weighed, and stored for analysis. Every 2 weeks 10% of the leaves were removed. The leaves were then dried, ground, and stored for analysis. In mid April 2001, leaves of the trees were removed and the total dry biomass determined.

Survey of clones at the Aokautere Nursery, Palmerston North

In mid April 2003 at the Aokautere Nursery, leaves were harvested from the stool beds of seven poplar clones and two willow clones, as well as from nearby pasture. All samples were taken within a 150 m radius. For each tree, approximately 100 g of fresh leaf material was taken from 1-year-old growth, with leaves being removed from the apex to the base of each shoot. For each poplar and willow clone, five trees were sampled. Five pasture samples were also taken. Table 2 shows a list of the species taken with their common and scientific names.

Soil was taken from the root zone of each tree or pasture plant that was sampled. Where sampled rows were adjacent, one soil sample was taken from an intermediate position. The soil at the Aokautere Nursery is a fine sandy loam (Manawatu series) with pH = 5.4, cation exchange capacity (CEC) = 13.4 cmol_c kg⁻¹, organic carbon content of 63 g kg⁻¹, and a resident Cd concentration of 0.3 μ g g⁻¹.



Fig. 1 Poplar sampling locations in the North Island, New Zealand.

Survey of 'Kawa' and 'Veronese' from locations around the North Island

Eight landowners from around the North Island (Fig. 1) sent in plant samples from hybrid poplars, as well as their associated soils from various locations around their properties. We were unable to identify

	Number	Common name	Scientific name
Pasture	1		Agrostis tenuis
Poplar	2	'Yeogi'	Populus alba × glandulosa
1	3	'Kawa'	Populus deltoides × yunnanensis
	4	'Veronese'	Populus deltoides × nigra
	5	'Shinsei'	Populus nigra × maximowiczii
	6	'Crow's Nest'	Populus euramericana × nigra
	7	Chinese poplar	Populus yunnanensis
	8	'Toa'	$Populus$ euramericana \times yunnanensis
Willow	9	'Booth'	Salix purpurea
	10	'Kinuyanagi'	Salix schwerinii

 Table 2
 Poplar, willow, and pasture species analysed in these experiments.

all the clones from the leaves that were sent, however, the majority were 'Kawa' (*Populus deltoides* \times *P. yunnanensis*) and 'Veronese' (*Populus deltoides* \times *nigra*). Plant ages ranged from 4 to 20 years. The soil types were not determined.

Sample preparation and elemental determination

The concentrations of trace elements in the soil solution from the Aokautere Nursery survey were determined as follows. Soils were dried and sieved to <0.5 mm, then 4 g portions were weighed into 50 ml centrifuge tubes, and 40 ml of 0.5 *M* CaCl₂ solution added. After agitation overnight, the samples were filtered and the solutions stored for analyses.

For the total trace element concentrations, approximately 0.2 g portions of dried, ground sawdust, soil, leaf, and stem samples were accurately weighed into 50 ml Erlenmeyer flasks. Concentrated nitric acid (10 ml) was added to each tube and the mixtures heated on a heating block until a final volume of about 3 ml was reached. The samples were then diluted to 10 ml using deionised water and stored in polyethylene containers. Elemental determinations for the lysimeter experiment were made using Inductively Coupled Plasma Emission Spectroscopy (ICPES). Elemental determinations for the survey of poplar and willow clones at Aokautere were made using Flame Atomic Absorption Spectroscopy (FAAS) for Cu, Fe, Mn, and Zn. Graphite Furnace Atomic Absorption Spectroscopy (GFAAS) was used for Co and Cd, and ICPES for B.

Data analysis

Data from the trace element concentration measurements were analysed using MINITAB. Fisher's least significant difference method was used to indicate significant difference between the trace element concentrations of different clones.

RESULTS AND DISCUSSION

Trace element accumulation by 'Toa' growing in lysimeters

The average leaf biomass production of the trees was 104, 183, and 329 g of dry leaves per tree for the low, medium, and high fertilisation treatments, respectively. The trees grown in the "low" fertilisation treatment showed visible signs of nutrient deficiency such as stunting and chlorosis. However, there were no significant differences in the leaf trace element concentrations between the different fertilisation regimes. This indicates that, at least for the trace elements tested, the macronutrient status of the tree has not affected the leaf trace element concentration. Nevertheless, the amount of trace element extracted per tree increased with increasing biomass production. Table 1 shows the average amounts of trace elements that leached from each lysimeter. The amount leached was 8–60 times more than the amount added with the Hoagland's solution in the "high" treatment. This indicates that the sawdust trace element concentrations decreased during the experiment.

Figure 2A–F shows the average leaf trace element concentrations for all the poplars in the greenhouse experiment. The concentration of Co in the leaves was below detection limits in all the trees and is not shown.

At the end of the season, leaves of the poplar 'Toa' clone had high concentrations of B, Cd, and Zn relative to values reported for pasture species (Underwood & Suttle 1999). Values of Cu, Fe, and Mn fell within the normal range for plants (Salisbury & Ross 1992). No conclusions regarding nutritional value can be drawn, however, from the total trace element concentrations of poplars grown in sawdust because the chemical composition of this material differs greatly from soil. Nevertheless, this experiment elucidated the change in leaf trace element concentration over time.

Except for Fe, there were significant changes for all elements with respect to time. Boron, Cd, Zn, and Mn were highly significantly or very highly significantly and positively correlated with time. Conversely, Cu was highly significantly and negatively correlated with time. Thus for B, Cd, Zn, and Mn, using tree material for fodder at the end of the growing season is likely to result in higher foliar concentrations. The converse is true for Cu. A possible explanation for the increases in foliar concentration for the trace elements studied is that they are transported in the xylem and thus accumulate in the leaves, which are a major water sink. The patterns observed here are similar to those observed by Laureysens et al. (2004) for European poplar varieties.

Trace element accumulation by poplars, willows and pasture from the Aokautere nursery and around New Zealand

Figure 3 illustrates the ability of some poplars and willows to take up significant amounts of Cd from soil compared to pasture. Willows, in particular, had

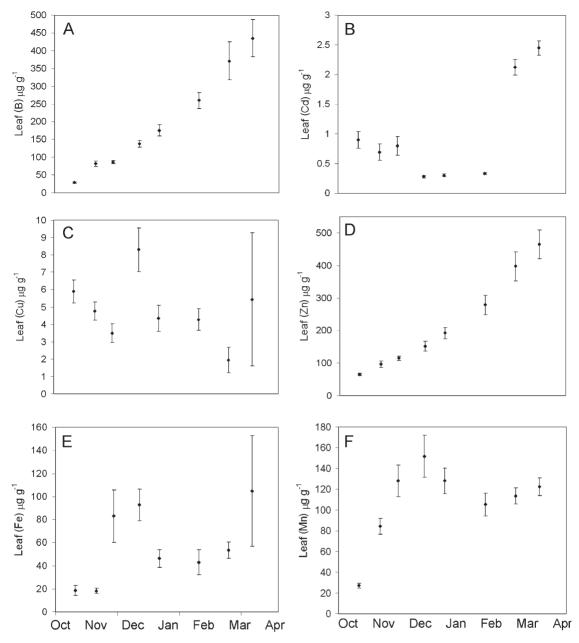


Fig. 2 A-F, Leaf trace element concentration in 'Toa' (*Populus euramericana* × *yunnanensis*) growing in lysimeters filled with sawdust. The error bars represent the SEM. Note the changed scale on the ordinates.

a leaf Cd concentration that was many times greater than pasture growing on the same soil.

soil. Table 3 shows the average values for the total and soluble trace elements measured in this study.

Chemical analyses of the soils revealed no significant differences in the soil properties sampled between clones, indicating that differences in plant uptake of trace elements are due to factors other than Given that the Cd concentration in the soils of the Aokautere Nursery $(0.1 \ \mu g \ g^{-1})$ is lower than the national average (0.44 $\mu g \ g^{-1}$), the Cd concentrations in poplar and willow trees used for soil conservation

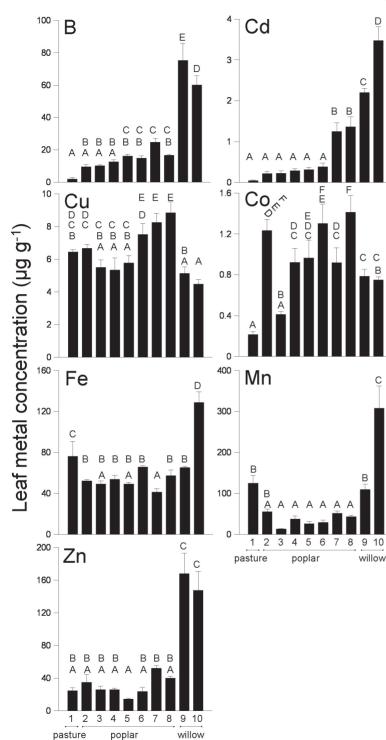


Fig. 3 Trace element accumulation in pasture, poplars, and willows from the Aokautere Nursery. Bars with the same letters are not significantly different. Table 2 identifies the species (1–10). The soil concentrations are shown in Table 3. The error bars represent the SEM.

and fodder purposes may well exceed the values shown in Fig. 3. Our North Island survey of poplars ('Kawa' + 'Veronese') confirmed this (Table 4). The average leaf Cd concentration was 1.8 μ g g⁻¹ from the sites around the North Island, compared to <0.5 μ g g⁻¹ at Aokautere. The average Cd concentration in leaves from the North Island survey is greater than the concentration (1 μ g g⁻¹) that has been shown to adversely affect livestock (Underwood & Suttle 1999). It is unclear, however, what adverse effect (if any) poplar and willow leaves may have if they only form a small part of the animal's diet.

The exposure of stock to Cd can be managed by planting fodder clones that take up lesser amounts. There is a clear demarcation between the willows, 'Toa', 'Chinese poplar', and the other varieties of poplar (Fig. 3). The aforementioned species should be avoided if the soil contains elevated levels of Cd, such as in pasture land that has been intensely fertilised with superphosphate over a long period.

Younger leaves had lower Cd concentrations than older leaves (Fig. 2). Therefore, leaves harvested later in the season (i.e., older) will contain higher amounts of Cd. Unfortunately, this is the time when poplars and willows are most valuable as fodder because they are often used to alleviate the effects of late summer droughts. Nevertheless, feeding out poplars before hay is required may lessen the stock's intake of Cd. Taking leaves towards the tips of the branches would also result in lower Cd exposure, although this may not be practical.

Leaf concentrations of B, Co, Cu, Fe, Mn, and Zn provided additional information about the potential fodder value of different poplar and willow clones (Fig. 3). Significant variation between species was found for all trace elements. These elements were

Table 3 Mean metal concentrations ($\mu g g^{-1}$) of the 30 soil samples taken from the Aokautere Nursery. Values in brackets are the SEM.

Element	Total concentration in soil (µg g ⁻¹)	Concentration in soil solution (µg ml ⁻¹)		
В	<0.5	< 0.5		
Cd	0.1 (0.0)	< 0.05		
Со	2.1 (0.1)	< 0.05		
Cu	5.6 (0.2)	0.14 (0.01)		
Fe	12644 (253)	0.95 (0.17)		
Mn	199.2 (4.3)	15.0 (0.9)		
Zn	36.4 (0.6)	0.6 (0.1)		

Table 4 Elemental concentrations ($\mu g g^{-1} dry weight$) in poplars ('Kawa' + 'Veronese') and soils from around the North Island. n.d. = not determined.

	Cd	Cu	Zn	Co	Fe	Mn
Soils $(n = 35)$						
Average	1.3	7.9	35	5.5	n.d.	292
SE	0.1	0.7	2.5	0.3	n.d.	36
Min.	0.1	0.6	14	2.4	n.d.	90.2
Max.	3.1	19.1	77	9.2	n.d.	856
Stems $(n = 13)$						
Average	1.4	6.5	43	0.5	42	66
SE	0.1	1.1	5.3	0.1	7.8	15.5
Min.	0.4	2.0	15	0.1	4	12
Max.	1.9	15.8	68	1.1	92	252
Leaves $(n = 37)$						
Average	1.8	8.3	165	4.6	132	195
SE	0.2	0.9	17.6	0.6	20.3	47.1
Min.	0.1	0.1	29	0.5	18	12
Max.	4.8	30.3	431	15.9	546	1539

	Sheep	Cattle
Dry matter intake (kg yr ⁻¹)	700	3000
Co intake (g yr ⁻¹) on normal (0.15 μ g g ⁻¹) pasture	0.105	0.45
Co intake $(g yr^{-1})$ on impoverished (0.06 $\mu g g^{-1})$ pasture	0.042	0.18
Poplar leaves (kg) required (@ 2 μ g g ⁻¹ Co) to make up shortfall	33	114

 Table 5
 A Co balance for sheep and cattle. All values are reported on a dry matter basis.

concentrated mostly in the leaves (Table 4). The leaf concentrations of B, Co, and to a lesser extent Zn, were much greater in the poplar and willow clones than in the pasture.

Boron is a non-essential element for animals. The leaf concentrations reported here (Fig. 3) are well below the value ($800 \ \mu g \ g^{-1}$) that has been shown to be toxic to livestock (Underwood & Suttle 1999). However, the results of the lysimeter experiment (Fig. 2A) showed that when B is a contaminant in the substrate, poplars and willows can accumulate high concentrations.

Cadmium toxicity affects both Mn and Zn metabolism in calves and sheep. Zinc consumption may alleviate Cd toxicity in animals (van Bruwaene et al. 1986). Therefore, poplars and willows with a high Zn concentration could be selected for fodder use to reduce Cd toxicity to stock. In addition, supplemental feeding of poplar and willow leaves to stock may alleviate Zn deficiency in areas with a low soil Zn concentration. Clark & Millar (1983) reported a Zn concentration of less than 5 μ g g⁻¹ in New Zealand pasture species, compared with the mean Zn concentration of 140 μ g g⁻¹, found in willow leaves (Fig. 3).

The Co concentrations (Fig. 3) of all clones except 'Kawa' were significantly greater than in pasture. Our survey of poplars in the North Island revealed even higher concentrations (Table 4). Poplar fodder may thus be used to alleviate Co deficiency, or so-called "bush sickness", on farms deficient in this element.

Pasture has an average Co concentration of about 0.15 μ g g⁻¹. Pasture on Co-deficient soil can have concentrations as low as 0.06 μ g g⁻¹ (Andrews 1966). To avoid deficiency, sheep and cattle require a pasture with dry matter Co concentrations of 0.11 and 0.08 μ g g⁻¹, respectively (Andrews et al. 1958; Clark & Millar 1983). Assuming Co concentrations in dry poplar leaves to be at least 2 μ g g⁻¹, which is half the value found in our North Island survey, then Co deficiency on impoverished soils could be

alleviated by feeding just 33 and 114 kg of poplar leaves (dry weight) to sheep and cattle (Table 5). This Co balance is summarised in Table 5. Our calculations assume that all the Co that the animal ingests is bioavailable.

Clone selection for optimal trace element nutrition

The results of these experiments indicate that willows should be avoided as stock fodder in areas known to have a high soil Cd concentration. However, willows provide high concentrations of the essential trace nutrients Mn and Zn. 'Yeogi' and 'Crow's Nest' poplars provide high amounts of Zn, Cu, and Co, with minimal Cd uptake. These appear the best varieties to plant on the basis of trace element nutrition.

CONCLUSIONS

Compared to pasture, poplars and willows accumulate high concentrations of some trace elements in their leaves. With the exception of Cd, this accumulation is beneficial to stock health. Poplar and willow leaves may be used as a Co or Zn supplement in areas that are deficient in these elements. Poplars and willows could be managed by clone selection and time of harvest so that stock are exposed to minimum amounts of Cd while still benefiting from high concentrations of essential trace elements in the leaves.

Further work is needed to determine the effect of soil type and metal concentration on trace element uptake by poplars. This could elucidate ways of reducing or promoting trace element uptake by using appropriate soil conditioners. For example, adding lime to fodder trees growing on Cd-contaminated soils would raise the soil pH, thus reducing the solubility and consequent plant uptake of this toxic element.

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