Prediction of cadmium removal from highly contaminated soils by phytoextraction with different switchgrass cultivars

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Abstract
The energy crop, switchgrass (*Panicum virginatum* L.), has been studied for phytoextraction of heavy metals in soil because of its high biomass production and moderate tolerance to heavy metals. The potential of switchgrass to remove heavy metals from contaminated soils is promising. The performance of phytoextraction of heavy metal by switchgrass plant from highly contaminated soils has never been discussed. In this study, the maximum removal of Cd from soil during harvest of aboveground tissues of switchgrass was predicted with the relationship between plant Cd concentration and dry weight of switchgrass. Two eco-type cultivars, Alamo and Blackwell, were used for illustration. And, two soil series (Lukang and Pinchen) with spiked Cd (0, 20, 60, and 120 mg Cd/kg soil) were used in pot experiments. The aboveground tissues of switchgrass were harvested after a growth period of three months, and then dry weight and plant Cd concentration were measured. The results showed that the two eco-types of switchgrass, Alamo and Blackwell, were showed different phytotoxic responses to Cd. The phytotoxicity thresholds of 63% reduction in plant Cd concentration (PT63) for Alamo and Blackwell were respectively 46.51 and 98.04 mg/kg. This suggested that Blackwell certainly had higher Cd tolerance than Alamo did. Nevertheless, the maximum phytoextraction of Cd by Alamo and Blackwell, respectively, 141.3 and 47.1 µg/pot were happening on plant Cd concentrations of PT63. Alamo thus had higher efficiency to remove Cd from soil than Blackwell did. Because the maximum phytoextraction of Cd by Alamo was much higher than that by Blackwell, Alamo would be the priority cultivar to be used for Cd removal from heavily contaminated soils.

Key Words
Heavy metal, phytoavailability, phytotoxicity, phytoremediation, energy crop.

Introduction

Among heavy metals, cadmium (Cd) is of special concern due to its relatively high mobility in soils and potential toxicity to biota at low concentrations. Cadmium is usually released into the environment via various industrial activities. Smelters, heating systems, metal-working industries and cement factories are the main contributors to Cd pollution. Increasing concentrations of Cd have been observed in agricultural soils due to long term application of phosphorous fertilizers and sewage sludge (Stephens and Calder 2005). Since Cd tends to adsorb to the topsoil, phytoextraction has been proposed as a low-cost technique to reclaim Cd contamination of soils (Cosio *et al.* 2006).

For efficient remediation, the phytoextraction process needs to take into account good biomass yields and heavy metal hyperaccumulation simultaneously (McGrath and Zhao 2003). Maxted *et al.* (2007) used the product of plant Cd and biomass yield weight to measure phytoextractable Cd by *Thalspi caerulescens* grown under various pH and organic carbon concentration of lowly contaminated soils. While high plant Cd accumulation results in decrease of biomass yield under highly contaminated soils, the phytotoxic symptoms will violate measurement of phytoextractable Cd. So far, there have been few studies to show phytoextraction of heavy metal in highly contaminated soils.

Switchgrass (*Panicum virgatum* L.), which is a perennial grass of importance as a forage and hay crop in soil conservation and as a potential source for biofuels, is a promising plant for phytoextraction of heavy metals (Entry and Watrud 1998; Shahandeh and Hossner 2000). Reed *et al.* (1999) reported that switchgrass was tolerant of Cd while grown in sand culture. Accumulation of Cd in switchgrass was dependent upon soil pH, Cd concentration and plant cultivars (Reed *et al.* 2002). Because switchgrass could yield high biomass (Sladden *et al.* 1991) and tolerate moderate levels of Cd in soils to accumulate Cd in aboveground tissues (Reed *et al.* 1999; Reed *et al.* 2002), the potential of switchgrass for Cd phytoextraction in contaminated soil is thus promising.

Currently, the performance of Cd phytoextraction by switchgrass plant under high contamination has never been discussed. In order to assess the potential of switchgrass used for phytoremediation of Cd contaminated soils, the maximum removal of Cd from soil during harvest of aboveground tissues should be figured out. The objective of the present study was to predict the phytoextraction of Cd by switchgrass in highly contaminated soils with a specified phytotoxicity model.

Methods

Soil characterization and preparation of Cd-spiked soils

Two soils, Pinchen and Lukang, were sampled from the agricultural lands in Taiwan. They were, respectively, classified into Typic Kandiudox and Aquic Dystrudept. Soil samples were air-dried and passed through a 2 mm sieve to be prepared for soil analysis and a pot experiment. The selected soil properties are listed in Table 1. The texture classes, which would be of concern for soil water regime management, are sandy loam for Pinchen and sandy clayey loam for Lukang, respectively. The pH value of Pinchen soil (pH = 4.6) is much lower than that of Lukang soil (pH = 7.4). Compared with Lukang soil, Pinchen soil also showed a lower CEC, OC and total Kjeldahl nitrogen. Air-dried soil samples were amended with four levels of Cd: 0, 20, 60 and 120 mg/kg, in the form of $CdCl_2$. Cd-spiked soils underwent three wetting-drying cycles at room temperature (25^{o}C). Each wetting-drying cycle was about 4 weeks. Each Cd-spiked soil sample after three wetting-drying cycles was then ground and passed through a 2 mm sieve to thoroughly mix for the pot experiment with switchgrass.

Pot experiments and plant growth and Cd measurements

Two switchgrass cultivars, Alamo and Blackwell, were used as the model plants in pot experiments. Alamo is a lowland eco-type which has high yields in a given environment (Cassida *et al.* 2005). Blackwell is an upland eco-type which usually shows a small response to drought stress (Stroup *et al.* 2003). Five seedlings of Alamo and Blackwell germinated for three weeks were transplanted into a pot (16.5 cm diameter × 14 cm height) with 1 kg Cd-spiked soil. The pot plants were grown in the greenhouse under natural light and the plant growth period was for four months. At harvest, aboveground tissues of plant were then collected by cutting the shoots at the pot soil surface. The harvested tissues were oven-dried at 65^{o}C for 72 h to measure the dry weight. The Cd concentration of each dried plant sample was then determined with an AAS (PerkinElmer AAnalyst 200) following a H_2SO_4 / H_2O_2 digestion procedure.

Prediction of Cd phytoextraction by switchgrass with a phytotoxicity model

An exponential decay model $f(x)$ was used to present plant dry weight, which decreased with increase of plant Cd concentration (x), to assess Cd toxicity of switchgrass:

$$f(x) = a \exp(- bx), \quad [1]$$

where the parameters a and b respectively indicate the maximum dry weight and the reduction rate of dry weight during increase of plant Cd concentration. Then, phytoextraction of Cd (w) could be expressed as the product of x and $f(x)$:

$$w = ax \exp(- bx). \quad [2]$$

Theoretically, there is a maximum value (w_{max}) if x is equal to $1/b$:

$$w_{\max} = \frac{a}{b}\exp(-1)\ . \quad [3]$$

Moreover, there is reduction of dry weight of about 63% when x is equal to $1/b$. The value of $1/b$ thus was identified as the phytotoxicity threshold of 63% reduction (PT63) and used to assess the plant tolerance to Cd.

Results

The effects of Cd-spiked soils on plant Cd concentration and dry weight are shown in Figure 1. Plant Cd concentration increased with increase of Cd level in the soil. However, dry weight was significantly decreased by increasing the levels of Cd in the soil. This suggested that Cd phytoavailability was dependent on Cd-contaminated levels and that phytotoxic symptoms of Cd in switchgrass plant were associated with an increase of Cd phytoavailability. In Figure 2, the effects of soil properties on plant Cd concentration and dry weight of switchgrass are shown. Switchgrass plants grown in Pinchen soil had a higher plant Cd concentration than those grown in Lukang soil (Figure 2(a)). Compared with the Lukang soil, Cd phytoavailability in the Pinchen soil was relatively high. This was because the values of pH, CEC and OC of the Pinchen soil were much lower than those of the Lukang soil. The low pH would raise the labile Cd of soil and the low CEC and OC would allow less sorption of Cd. More serious damage due to Cd phytotoxicity thus occurred when switchgrass plants were grown on the Pinchen soil (Figure 2(b)).

In Figure 3, relationships between plant Cd concentration and dry weight were expressed to show the Cd phytotoxicity of switchgrass. The observations for the Pinchen soil were clustered and obviously lower than those for the Lukang soil. This indicated that the phytotoxicity of Cd was more pronounced in the Pinchen soil than in the Lukang soil. Phytotoxicity models as Eq. [1] for Alamo and Blackwell cultivars were obtained by using best-fit regression. For the Alamo cultivar, a = 8.45 g/pot and b = 0.022, and for the Blackwell cultivar, a = 1.28 g/pot and b = 0.010. The maximum dry weight of Alamo was obviously higher than that of Blackwell; however, the reduction rate of dry weight of Blackwell was much lower than that of Alamo. By contrast, Alamo did not only have higher biomass production in aboveground tissues but also showed more dramatically Cd toxicity symptoms. Moreover, the PT63 values in plant Cd concentration for Alamo and Blackwell were, respectively, 46.51 and 98.04 mg/kg. This revealed that Cd tolerance of Blackwell was higher than that of Alamo.

Moreover, plant Cd concentrations of PT63 could indicate a desirable Cd phytoavailability to meet the maximum Cd phytoextraction (w_{max}), as shown Eq. [3]. The w_{max} values for Alamo and Blackwell at PT63 were, respectively, 141.3 and 47.1 µg/pot. This suggested that Alamo had a higher efficiency for Cd removal from soil than Blackwell did. For Cd accumulation by plants during phytoextraction, Blackwell might be suitable for a soil with highly available Cd and Alamo would be suitable in a soil with relatively low Cd availability.

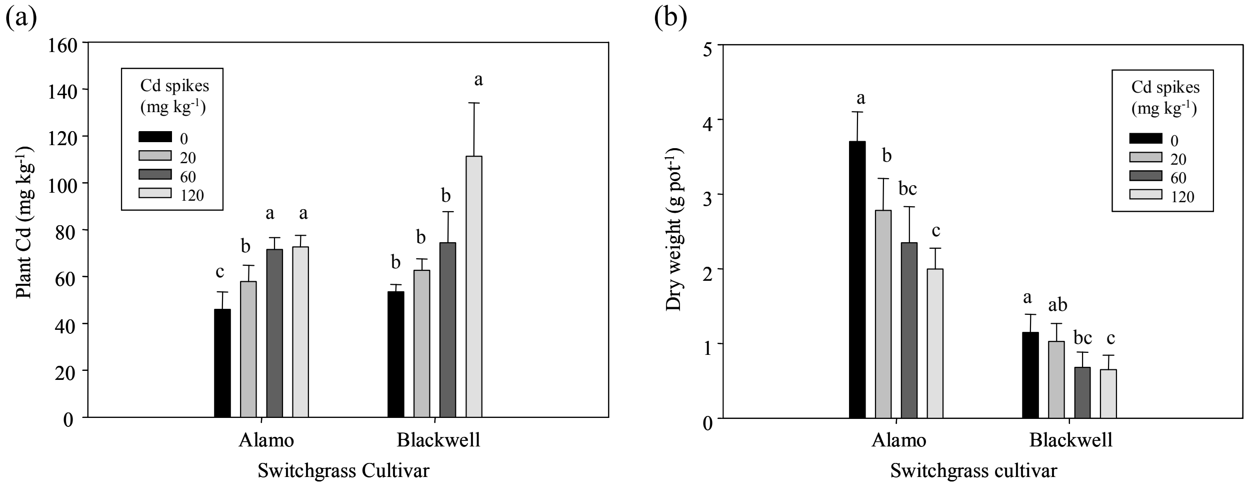

Figure 1. Effects of the four levels of Cd (0, 20, 60, and 120 mg/kg) in soils on (a) plant Cd concentration and (b) dry weight of switchgrass (Alamo or Blackwell) grown in the soils. Error bar is standard error of the mean for a given Cd level. Significantly different effects on switchgrass at the 0.05 level according to the LSD test are labeled with different letters.

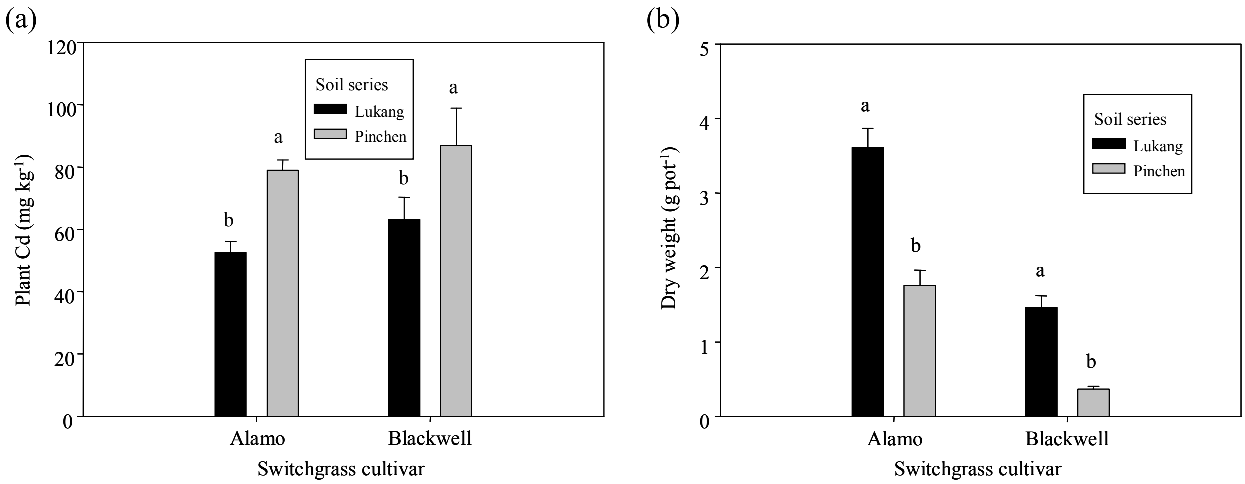

Figure 2. Effects of the two soil series (Lukang and Pinchen) on (a) plant Cd concentration and (b) dry weight of switchgrass (Alamo or Blackwell) grown in Cd-spiked soil. Error bar is standard error of the mean for a given soil series. Significantly different effects on switchgrass at the 0.05 level according to the LSD test are labeled with different letters.

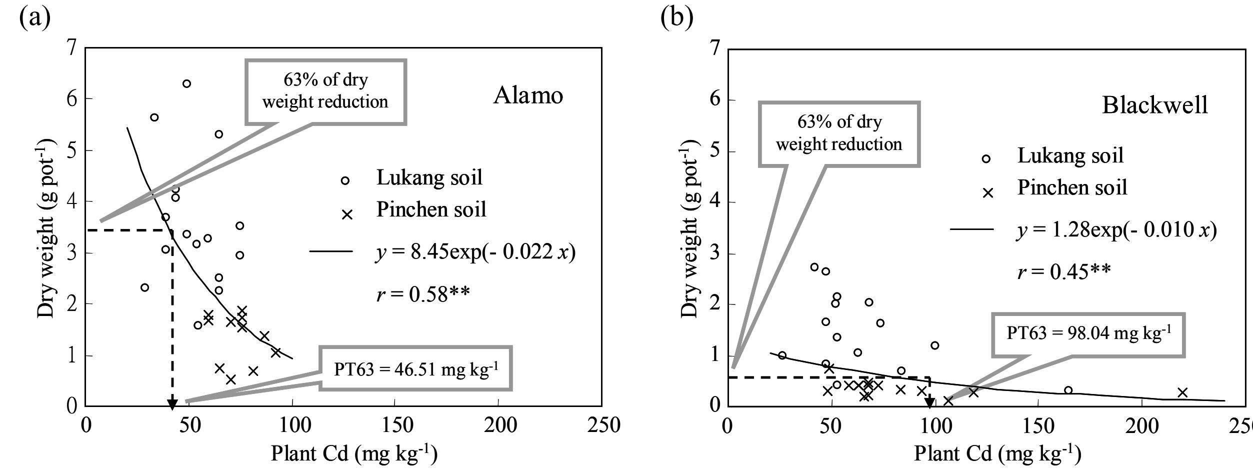

Figure 3. Relationships between plant Cd concentration and dry weight for the switchgrass cultivars, (a) Alamo and (b) Blackwell, grown in Cd-spiked soils. Plant Cd concentrations, with phytotoxicity thresholds of 63% dry weight reductions (PT63), for Alamo and Blackwell, respectively, are denoted. Best-fit regression models are represented by solid lines and with correlation coefficients *r* significant at *P* < 0.01 (**).

Conclusion

The two eco-types of switchgrass, Alamo and Blackwell, showed different phytotoxic responses to Cd. The PT63 values for Alamo and Blackwell were, respectively, 46.51 and 98.04 mg/kg. This revealed that Blackwell had higher tolerance to Cd than Alamo did. Nevertheless, the maximum phytoextraction of Cd by Alamo (141.3 µg/pot) was much higher than that by Blackwell (47.1 µg/pot). Alamo had higher efficiency for Cd removal from soil than Blackwell did. Therefore, Alamo switchgrass is suggested to be the priority plant for phytoextraction of Cd in heavily contaminated soils.

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