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**Long-term Field Phytoextraction of Zinc/Cadmium Contaminated Soil by *Sedum plumbizincicola* under Different Agronomic Strategies**

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# ABSTRACT

In two long-term field experiments the zinc (Zn)/cadmium (Cd) hyperaccumulator *Sedum plumbizincicola* (*S. plumbizincicola*) was examined to optimize the phytoextraction of metal contaminated soil by two agronomic strategies of intercropping with maize (*Zea mays*) and plant

densities. Soil total Zn and Cd concentrations decreased markedly after long-term phytoextraction. But shoot biomass and Cd and Zn concentrations showed no significant difference with increasing remediation time. In the intercropping experiment the phytoremediation efficiency in the treatment “*S. plumbizincicola* intercropped with maize” was higher than in *S. plumbizincicola* monocropping, and Cd concentrations of corn were below the maximum national limit. In the plant density experiment the phytoremediation efficiency increased with increasing plant density and 440,000 plants ha<sup>-1</sup> gave the maximum rate. These results indicated that *S. plumbizincicola* at an appropriate planting density and intercropped with maize can achieve high remediation efficiency to contaminated soil without affecting the cereal crop productivity. This cropping system combines adequate agricultural production with soil heavy metal phytoextraction.

**Key words**

heavy metal, phytoremediation, long term, intercropping, plant densities

## 1 INTRODUCTION

Soil contamination represents a risk to human health in various ways including contamination of food crops grown in polluted soils and contamination of groundwater or surface waters used as sources of drinking water or direct ingestion of contaminated soil (Muchuweti *et al.* 2006; Guney *et al.* 2010). Remediation of contaminated agricultural soils is therefore important to prevent the movement of potentially toxic methods via the human food chain (Wang *et al.* 2001). In recent years the use of repeated phytoextraction has attracted considerable interest (Epelde *et al.* 2008; Jadia and Fulekar 2009). The strategy of phytoextraction using metal hyperaccumulator plants has therefore been proposed as a potential technique for the decontamination of metal-polluted soils (Baker *et al.* 1994; Brown *et al.* 1995; Salt *et al.* 1995). The technique uses plants that hyperaccumulate potentially toxic elements in the aboveground biomass which can be harvested and removed from polluted soils. The results of field experiments indicate that metal phytoextraction using *Thlaspi caerulescens* may be used to clean up soils moderately contaminated by Cd (McGrath *et al.* 2006; Japenga *et al.* 2007; Koopmans *et al.* 2008).

*Sedum plumbizincicola* (*S. plumbizincicola*) also has a remarkable capacity to extract zinc (Zn) and Cd from polluted soils, and pot experiments have indicated the potential of *S. plumbizincicola* for Zn and Cd phytoextraction (Wu *et al.* 2006; Jiang *et al.* 2010). Although *S. plumbizincicola* shows high ability to accumulate Zn and Cd, phytoextraction is also affected by the availability of heavy metals in the soil. Numerous factors can affect the remediation efficiency of *S. plumbizincicola* and systematic field experiments are required to study

agronomic measures for the optimisation of plant yield and of heavy metal accumulation for efficient extraction of metals from soils (Zhuang *et al.* 2009). Agronomic measures include fertilizer application, control of planting density, harvesting methods, and intercropping systems that can influence the phytoavailability of potentially toxic elements in soils (Huang *et al.* 2012). The planting of a hyperaccumulator species together with an agricultural crop species (co-cropping) may be a useful option (Wu *et al.* 2007). Maize (*Zea mays*) is an important agricultural crop worldwide with high yielding potential and is thus of interest in intercropping systems for soil metal phytoremediation (Wojcik and Tukiendorf 2005; Wei *et al.* 2011).

Most of the published studies on *S. plumbizincicola* phytoextraction have been based on short-term remediation *in-situ* field phytoremediation or on pot experiments (Jiang *et al.* 2010; Wu *et al.* 2012; Li *et al.* 2014a; Li *et al.* 2014b). More long-term field experiments are thus required to support agricultural technology and extension work. In the present study two *in-situ* field plot experiments investigated intercropping with maize and sorghum versus monocropping and the role of planting density on heavy metal phytoextraction. Changes in soil heavy metals after repeated phytoextraction and the relationship between metals in the soil and metal uptake by the hyperaccumulator *S. plumbizincicola* were studied. The aim was to determine a suitable planting density of *S. plumbizincicola* for safe agricultural production of maize and sorghum and simultaneous phytoextraction of heavy metals from a contaminated soil.

## 2 MATERIALS AND METHODS

### 2.1 Soil characterization and field experiment design

The field site is located in the suburbs of Hangzhou city, Zhejiang province, east China. The soil is a Typic Agriudic Ferrosol. The climate is moist monsoon with an annual precipitation of approximately 1425 mm and a mean temperature of 16 °C. Rainfall occurs mainly from September to June with a maximum during May and June each year. Agriculture is the principal land use. The site is adjacent to a copper smelting factory that was operational from 1989 to 2000. Large quantities of metals have been emitted to the atmosphere in the form of fly ash and then deposited on soils near the smelter because the unregulated factory had no safe working practices or equipment for dust removal. So, the neighbouring area was suffered by Cd, Zn, Cu and Pb pollution from flying ash from the factory for many years. The locations of the field experiment plots of this study were about 30 to 50 m from the pollution source, so the metal pollution levels of each plot was varied markedly with the distance to the copper smelter.

The soil pH (in H<sub>2</sub>O) was 7.24, the organic carbon content was 29.1 g kg<sup>-1</sup>, and the cation exchange capacity (CEC) was 11.8 cmol (+) kg<sup>-1</sup>. Total N, P, and K were 2.21, 0.22, and 22.9 g kg<sup>-1</sup>, respectively. Available N was 105 mg kg<sup>-1</sup>, Olsen-P was 6.70 mg kg<sup>-1</sup>, and NH<sub>4</sub>OAc-extractable K was 160 mg kg<sup>-1</sup>.

### 2.1.1 Field experiment: phytoextraction using intercropping

The phytoextraction field experiment was carried out from 2006 to 2014 over eight successive crops. There were three planting treatments, namely monocropped maize or sorghum (*Sorghum bicolor*) with harvest of sorghum from 2007 to 2009 and of maize from 2010 to 2014, the monocropped hyperaccumulator *S. plumbizincicola*, and maize (or sorghum) intercropped with *S. plumbizincicola*. There were four replicates of each treatment giving a total of 12 plots. Each plot was 6 m long and 5 m wide and was fertilized with 150 kg urea and 225 kg compound

fertilizer (15% N: 15% P<sub>2</sub>O<sub>5</sub>: 15% K<sub>2</sub>O) ha<sup>-1</sup>. The plots were fertilized before the next growth season with the same fertilizers as described above.

The sampling depth was the 0 - 15 cm arable layer and the soil samples were retained for determination of heavy metals in 2008 and 2014. Plants were harvested once a year. The shoots of the crops were harvested in June 2007, April 2009, June 2010, July 2011, July 2012, July 2013, and July 2014. The experiment started in 2006 when the Cd and Zn concentrations were 4.58±0.34 to 2.98±0.47 mg kg<sup>-1</sup> and 2527±155 to 1561±106 mg kg<sup>-1</sup>, respectively. According to the Chinese Soil Environmental Standards (GB 1995-15618), both Cd and Zn in soil were much higher than the permissible levels for agricultural soil (0.6 and 250 mg kg<sup>-1</sup>, respectively).

### 2.1.2 Field experiment: effect of planting density on phytoextraction efficiency

The phytoextraction field trial was carried out from 2008 to 2014 over six successive crops. There were four planting densities, namely 110 (D11), 250 (D25), 440 (D44), and one thousand seedlings per hectare. Each treatment had three replicates and each plot was 6 m long and 2 m wide. Seedlings of *S. plumbizincicola* were transplanted on 14 May 2007. The soil was then fertilized with 150 kg urea and 225 kg compound fertilizer (15% N: 15% P<sub>2</sub>O<sub>5</sub>: 15% K<sub>2</sub>O) ha<sup>-1</sup>. On 20 April 2008 the field soil was fertilized with 208 kg urea and 417 kg compound fertilizer ha<sup>-1</sup>. The soils were fertilized before the start of the next plant growth season with the same fertilizers as described above.

Plants were harvested once per year. The sampling depth was 0 - 15 cm since the cultivation depth in this region is 15 cm. The soils were retained for determination of heavy metals in 2008, 2009, 2013, and 2014. The shoots of the first, second, third, fourth, fifth and sixth crops were harvested in June 2008, April 2009, June 2010, July 2011, July 2012, July 2013, and July 2014.

The experiment was started in 2007; the Cd and Zn concentrations were  $3.04 \pm 0.11 \text{ mg kg}^{-1}$  and  $1299 \pm 96 \text{ mg kg}^{-1}$ , respectively.

## 2.2 Determination of metals in soil and plant samples

Soil total heavy metal concentrations were determined by atomic absorption spectrophotometry (Varian SpectrAA 220FS, 220Z; Varian, Palo Alto, CA) after digestion of 0.25g samples with 12 ml of HCl: HNO<sub>3</sub> (4: 1, v/v). Plant samples (0.25 g) were digested using a mixture of 6 ml HNO<sub>3</sub> and 4 ml HClO<sub>4</sub> and the metals were also determined by AAS. Replicate samples, blanks, and a certified reference material (GBW07401, provided by the Institute of Geophysical and Geochemical Exploration, Langfang, Hebei province, China) were included in all analyses for quality control. All chemicals used were of analytical reagent grade and the reference material results obtained by the methods described above were within the certified ranges.

## 2.3 Statistical analysis

Data were analysed by one-way analysis of variance using the SPSS for Windows version 13.0 software package. The data are presented as mean  $\pm$  standard deviation of the mean (SD).

# 3 RESULTS

## 3.1 Phytoextraction using intercropping

### 3.1.1 Plant biomass and metal concentrations in each crop

The plant biomass of each crop in the repeated phytoextraction sequence is listed in Table 1. From 2007 to 2009, irrespective of monocropping or intercropping, there were no significant



differences in sorghum biomass. And also the biomass of maize between the years of 2010 and 2014 were no significant differences. From 2007 to 2014 there was no discernible reduction in shoot biomass of plant species either in monoculture or intercropping. *S. plumbizincicola* grew well in monocropping and intercropping except in 2007 the biomass was lower than in other years. This was mainly due to the inappropriate seedling transplanting time and the high temperatures experienced during the seedling growth period. And there was no significant difference in the biomass of *S. plumbizincicola* between monoculture and intercropping treatments.

The concentrations of Zn and Cd in the plant shoots are shown in Tables 2 and 3. Zinc and Cd concentration in *S. plumbizincicola* were much higher than other plants. With increasing phytoextraction period, there was no significant difference in plant shoot Zn and Cd concentrations with exception of the lower Cd concentrations in *S. plumbizincicola* in 2007 than in other years, which might because of the poor growth of this plant in year 2007. According to the Chinese National Food Quality Standard for Cd in grain is  $0.1 \text{ mg kg}^{-1}$  (GB 2762-2012), the Cd concentrations in maize and sorghum grains in the treatments were below the limit.

### 3.1.2 Zinc and cadmium concentrations in soil during remediation

The Zn and Cd concentrations in soils after eight crops of phytoextraction are shown in Table 4. Because of the high heterogeneity of metal concentrations among the plots, metal concentrations in each plot were separately listed in the table. Both soil Zn and soil Cd decreased markedly after remediation. The average largest decreases occurred in the maize/*Sedum* intercropping treatment and after eight years' phytoextraction (from 2006 to 2014) the soil Cd and Zn were decreased from  $3.50 \pm 0.49$  and  $1799 \pm 215 \text{ mg kg}^{-1}$  (in 2006) to  $0.55 \pm 0.44$  and

1465±228 mg kg<sup>-1</sup> (in 2014), respectively. Compared to the metal concentration in 2006, after eight crops of repeated remediation the Zn concentrations in the maize monocropping, *Sedum* monocropping and maize/*Sedum* intercropping in 2014 decreased by 7.2, 13.7 and 18.8%, respectively; and the Cd concentration decreased by 40.9, 71.9 and 85.4%, respectively. The phytoremediation efficiency of Zn and Cd by *S. plumbizincicola* intercropped with maize was higher than by the *S. plumbizincicola* monoculture.

### 3.2 Phytoextraction efficiency experiment

#### 3.2.1 Plant biomass and metal concentrations in each crop

The *S. plumbizincicola* plant biomass in the repeated phytoextraction sequence is shown in Fig. 1. There were significant differences in shoot biomass between the treatments. Shoot biomass in treatment D44 was higher than in D11. Plant Zn and Cd concentrations appeared to increase gradually with increasing plant density but there were no significant differences among phytoextraction treatments. As the phytoextraction time increased the shoot Zn and Cd concentrations in plants showed no differences except for 2010 to 2012 and this was mainly due to the low plant biomass.

#### 3.2.2 Soil Zn and Cd concentrations during remediation

Table 5 shows the Zn and Cd concentrations in the soils after seven crops of phytoextraction. The Cd and Zn concentrations in treatments D11, D25, and D44 decreased markedly from 3.04 mg kg<sup>-1</sup> to 0.82, 0.56, and 0.37 mg kg<sup>-1</sup>, and from 1299 mg kg<sup>-1</sup> to 1037, 978, and 934 mg kg<sup>-1</sup>, respectively. With increasing remediation time the soil Zn and Cd concentrations declined significantly under the different planting densities, and the soil Zn and Cd concentrations decreased gradually with increasing plant density.

Compared with the soil before phytoextraction, after seven years of *S. plumbizincicola* phytoremediation the soil Cd concentrations in treatments D11, D25, and D44 were all lower than the national soil environmental quality standard levels (Cd 0.6 mg kg<sup>-1</sup>). Compared to soil without remediation the Zn concentrations in treatments D11, D25, and D44 decreased by 20.2, 24.7 and 28.1%, respectively, and the Cd concentration decreased by 73.0, 81.6 and 87.8%. Thus, the phytoremediation efficiency increased with increasing plant density, and the optimum planting density was 440,000 plants ha<sup>-1</sup>.

## 4. DISCUSSION

### 4.1 Differences between long-term field remediation experiments and short-term pot experiments

*S. plumbizincicola* has a remarkable capacity to extract Zn and Cd from contaminated soils and field experiments have indicated the potential of *S. plumbizincicola* for Cd and Zn phytoextraction (Wu *et al.* 2006). The study by Jiang *et al.* (2010) also provides evidence for the potential use of *S. plumbizincicola* for repeated phytoextraction in greenhouse experiments. In this study, the capacity of *S. plumbizincicola* was examined in field condition during the long-term phytoextraction. There was no discernible reduction in plant shoot biomass in monoculture or intercropping treatments during the periods of long-term phytoextraction, indicating that there were no negative continuous cropping effects occurring over time with repeated cropping in this study. Significantly higher Cd concentrations in *S. plumbizincicola* shoots were found in the field microcosm experiment compared to the pot experiment the same

soils used as this study (Li *et al.* 2014a). Meanwhile the Cd concentration in soil planted with *S. plumbizincicola* decreased significantly after remediation in field experiments (Table 4) and this is similar to the changes observed in a glasshouse long-term remediation experiment (Li *et al.* 2014a). This discrepancy may be due to the limited and discontinuous soil volume in pots. With resulting differences in the distribution of roots, soil temperature where the roots do occur, and diurnal changes in water content (Watts 1975; Barber *et al.* 1988). And Barber *et al.* (1988) also reported that soil temperature and water content had significant effects on root growth. Moreover, under field conditions the soil is a continuum that can supply soluble metals soon after absorption by plants from the soil solution, and plant growth cycle in field is longer than the pot experiment, leading to a substantial difference in plant growth conditions between pot and field experiments.

Successful phytoextraction requires that the polluted medium is cleaned to a level that complies with environmental regulations and from an economic viewpoint this should be achieved at a lower cost than an alternative technology or the cost of inaction (Robinson *et al.* 2003). However, despite intensive research on the subject in the last decade, very few field studies or commercial operations that demonstrate successful phytoextraction have been realized (Robinson *et al.* 2007). In the present study the soil Zn and Cd concentrations occurred in the maize/*Sedum* intercropping treatment after eight crops of phytoextraction declined from  $3.50 \pm 0.49$  to  $0.55 \pm 0.44$  mg kg<sup>-1</sup>. After seven crops of phytoremediation by *S. plumbizincicola* with different planting densities the soil Cd concentration was  $0.82 \pm 0.32$ ,  $0.56 \pm 0.31$ , or  $0.37 \pm 0.74$  mg kg<sup>-1</sup> respectively, and reached the third level of the National Soil Environmental Quality Standard (Cd 0.60 mg kg<sup>-1</sup>).

#### 4.2 Fate of heavy metals in soil during phytoremediation under field conditions

After long-term phytoextraction, soil metal showed the apparent decreases in both filed experiments of this study. The decreases in soil Cd and Zn in the *S. plumbizincicola* monoculture, maize monoculture and *S. plumbizincicola* intercropped with maize were 3.76 to 6.65 kg ha<sup>-1</sup> and 1069 to 1238 kg ha<sup>-1</sup>, respectively. And also the decline in soil Cd and Zn under different plant densities were 2.90 to 3.57 kg ha<sup>-1</sup> and 679 to 886 kg ha<sup>-1</sup>, respectively (Table 6). Metal uptake by remediating plant could be one of important reasons for soil metal decrease. The largest amounts of Cd and Zn taken up were 4.07 and 208 kg ha<sup>-1</sup> in the intercropping treatment, respectively. The largest plant Cd and Zn uptakes were 3.59 and 290 kg ha<sup>-1</sup> in the plant density experiment (D44). However, when comparing the metal uptake by plant with the soil metal decrease after phytoextraction, it could be found that the remediation efficiency calculated by the change in the concentration of heavy metals in the soil was higher than by the aboveground plant parts. One of reasons may be due to non-uniformity of soil samples collected. *S. plumbizincicola* was harvested when the soil samples were collected from the top 15 cm of the soil profile. *S. plumbizincicola* is a shallow rooting species and the heavy metals taken up by the plant aboveground parts may be concentrated in the top 10 cm of the soil profile, resulting in a marked depletion of heavy metals in the surface soil. In addition, heavy metals in the soil were controlled not only by plant uptake but also by surface runoff or leaching down the profile and to the atmosphere via dust (El Khalil *et al.* 2008). Accumulation of heavy metals in soils is likely to result in increased export of heavy metals to waters. The contents and fractions of heavy metals in soils directly influence the transport of the metals. Studies have also shown that heavy metals primarily accumulate in surface soil and are readily affected by rain, leading to an increasing

heavy metal loading in runoff (Zhang *et al.* 2004). It was therefore not unexpected that the decline in the amount of heavy metals in the soil was much greater than the uptake by aboveground plant parts.

### 4.3 Combined agricultural production and soil remediation

Hyperaccumulation with intercropping of crop species will allow simultaneous remediation of contaminated soils and production of agricultural goods and will optimise economic efficiency. This is particularly well suited to conditions of limited land resources and widespread soil pollution. Some studies demonstrated that co-cropping the hyperaccumulator *Sedum alfredii* with maize increased Zn and Cd phytoextraction (Hei *et al.* 2007; Jiang *et al.* 2013). Intercropping *S. plumbizincicola* with wheat in a wheat-rice rotation can improve the phytoremediation of heavy metal contaminated soil and reduce the food chain risk of rotated rice (Zhao *et al.* 2011). In the present experiment when *S. plumbizincicola* was intercropped with maize, the decreases in Cd and Zn were from 3.50 to 0.54 and 1799 to 1465 mg kg<sup>-1</sup>, respectively. Compared to the soil before phytoextraction, the Zn concentration from maize monocropping, *Sedum* monocropping and maize/*Sedum* intercropping decreased by 7.2, 13.7 and 18.8%, respectively, and Cd decreased by 40.9, 71.5 and 85.5%. The Chinese National Food Quality Standard for Cd in grain is 0.2 mg kg<sup>-1</sup> (Chinese Ministry of Health, 2012). Accordingly, the grain Cd concentrations of maize and sorghum in our experiment were below the National standard. In addition, under the field test conditions *S. plumbizincicola* grew well in soils with long-term with heavy metal contamination. Choosing an appropriate plant density can improve environmental factors such as light, water and heat utilization, leading to increased shoot biomass of *S. plumbizincicola* and shortening the time required for phytoremediation. In the planting density phytoextraction

experiment, with planting density increasing from  $1.1 \times 10^5$  (D11) to  $4.4 \times 10^5$  (D44) plants ha<sup>-1</sup>, Cd and Zn remediation efficiencies were 20.2, 24.7 and 28.1%, and 73.0, 81.6 and 87.8%, respectively. The remediation efficiency in D44 was the maximum. Appropriate planting density will benefit the Cd and Zn uptake by the aboveground parts of *S. plumbizincicola* and shorten the phytoremediation period. We can therefore use *S. plumbizincicola* accompanied by an appropriate planting density intercropping system with maize to increase the efficiency of remediation of contaminated soil and maintain normal agricultural production so as to achieve the dual purpose of simultaneous agricultural production and soil remediation.

## 5 CONCLUSIONS

Phytoextraction of metal-contaminated soils is a long-term process during which both soil Zn and soil Cd decrease markedly. In the *in-situ* phytoremediation experiment *S. plumbizincicola* intercropping with an agricultural crop not only remediates contaminated soil but also provides agricultural products. The phytoremediation efficiency of *S. plumbizincicola* increased with the increasing planting density. Therefore, *S. plumbizincicola* accompanied by an appropriate plant density, and intercropped with maize, can both increase the efficiency of remediation of contaminated soil and maintain normal agricultural production, so as to achieve the purpose of simultaneous agricultural production and soil remediation.

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**Table 1** Plant shoot biomass in the different cropping treatments (ton ha<sup>-1</sup>)

	Treatment		2007	2008	2009	2010	2011	2012	2013	2014
Maize or Sorghum	Sorghum	Straw	7.03±0.57a	6.54±0.52a	6.88±0.22a	-	-	-	-	-
Monocropping		Grain	4.79±0.38a	3.21±0.26bc	3.41±0.30b	-	-	-	-	-
	Maize	Straw	-	-	-	2.88±0.89a	2.11±0.11ba	2.86±0.50a	2.68±0.55a	2.52±0.18a
		Grain	-	-	-	3.13±0.23ab	2.45±0.47b	2.33±0.47b	3.41±0.38a	3.71±0.24a
Maize/Sorghum-Sedum	Sorghum	Straw	6.09±0.19a	4.71±0.23b	5.71±0.42a	-	-	-	-	-
Intercropping		Grain	6.23±0.24a	4.55±0.46b	5.55±0.58a	-	-	-	-	-
	Maize	Straw	-	-	-	3.71±0.34a	3.25±0.52a	2.98±0.33a	3.22±0.50a	3.13±0.27a
		Grain	-	-	-	4.11±0.17bc	4.31±0.46bc	5.88±0.35a	4.55±0.47b	3.62±0.42c
	Sedum		1.83±0.19c	3.62±0.42b	5.35±0.13a	6.01±0.28a	5.15±0.47ab	5.41±0.11ab	4.43±0.08b	4.25±0.08b
Sedum Monocropping	Sedum		1.38±0.32c	3.47±0.22b	5.21±0.46a	6.33±0.44a	5.78±0.69a	5.22±0.25ab	4.34±0.20b	4.15±0.47b

Values are means ± SD. Different letters in each row indicate the significant differences between years ( $p < 0.05$ ).

**Table 2** Zinc concentrations in plant shoots in the different cropping treatments (mg kg<sup>-1</sup>)

	Treatment		2007	2008	2009	2010	2011	2012	2013	2014
Maize or Sorghum	Sorghum	Straw	434±60a	399±33a	400±23a	-	-	-	-	-
Monocropping		Grain	58.0±6.0a	41.0±3.0b	43.0±7.0b	-	-	-	-	-
	Maize	Straw	-	-	-	299±40ab	345±29a	229±49b	277±24ab	300±54ab
		Grain	-	-	-	40.0±5.0a	38.0±4.0a	39.0±6.0a	40.0±11.0a	31.0±1.0a
Maize/Sorghum-Sedum	Sorghum	Straw	262±26a	291±14a	280±19a	-	-	-	-	-
Intercropping		Grain	43.0±5.0a	28.0±5.0b	38.0±6.0a	-	-	-	-	-
	Maize	Straw	-	-	-	291±56a	221±13ab	169±37b	234±35ab	264±19a
		Grain	-	-	-	38.0±3.0ab	34.0±4.0abc	42.0±6.0a	32.0±2.5bc	28.0±3.0c
	Sedum		5561±251a	5200±521a	5360±246a	4912±356a	4996±220a	5658±846a	5768±393a	5297±721a
Sedum Monocropping	Sedum		6680±778a	5500±456b	5013±321b	4617±549b	4843±241ab	5351±840ab	5649±210a	5208±329ab

Values are means ± SD. Different letters in each row indicate the significant differences between years (p<0.05).

**Table 3** Cadmium concentrations in plant shoots in the different cropping treatments (mg kg<sup>-1</sup>)

Treatment			2007	2008	2009	2010	2011	2012	2013	2014
Maize or Sorghum	Sorghum	Straw	3.80± 0.25a	2.83± 0.20b	2.76± 0.26b	-	-	-	-	-
Monocropping		Grain	0.14± 0.02a	0.09± 0.05ab	0.05± 0.00b	-	-	-	-	-
	Maize	Straw	-	-	-	1.83± 0.20a	1.65± 0.21a	1.50± 0.83a	1.40± 0.42a	2.17± 0.09a
		Grain	-	-	-	0.05± 0.02ab	0.04± 0.01ab	0.08± 0.05a	0.03± 0.01ab	0.02± 0.01b
Maize/Sorghum-Sedum	Sorghum	Straw	2.33± 0.33a	1.83± 0.57ab	1.33± 0.37b	-	-	-	-	-
Intercropping		Grain	0.07± 0.03a	0.07± 0.04a	0.07± 0.02a	-	-	-	-	-
	Maize	Straw	-	-	-	1.31± 0.58ab	1.45± 0.20ab	0.80± 0.23b	1.06± 0.31ab	1.82± 0.32a
		Grain	-	-	-	0.02± 0.01a	0.06± 0.02a	0.09± 0.06a	0.02± 0.01a	0.03± 0.01a
	Sedum		40.4± 5.1b	102±1 4a	100±1 4a	112±1 6a	98.0± 28.0a	132±2 2a	112±3 2a	114±6 4a
Sedum Monocropping	Sedum		76.5± 10.5b	155±1 6a	140±2 1a	156±5 2ab	162±4 5ab	198±8 0a	115±2 8b	110±3 2b

Values are means ± SD. Different letters in each row indicate the significant differences between years (p<0.05).



**Table 4** Total zinc and cadmium concentrations in soils in the different treatments (mg kg<sup>-1</sup>)

Treatment	Zn			Cd		
	2006	2008	2014	2006	2008	2014
Maize monocropping	2527±155	2250±230	2327±132	4.58±0.34	4.66±0.22	2.86±0.25
Maize monocropping	2140±200	2035±150	2011±210	4.55±0.42	4.31±0.36	3.13±0.36
Maize monocropping	1982±176	1964±330	1822±163	3.76±0.58	3.46±0.77	2.23±0.17
Maize monocropping	1854±136	1892±89	1730±254	3.72±0.16	3.79±0.21	1.71±0.11
<i>Sedum</i> monocropping	2406±101	2093±189	2178±56	4.50±0.24	4.01±0.89	2.23±0.09
<i>Sedum</i> monocropping	1789±88	1754±21	1574±77	3.59±0.22	3.30±0.96	1.07±0.13
<i>Sedum</i> monocropping	1762±96	1684±112	1514±34	3.22±0.14	3.25±0.35	0.65±0.03
<i>Sedum</i> monocropping	1762±58	1656±156	1497±28	3.07±0.35	2.97±0.74	0.44±0.02
Maize/ <i>Sedum</i> intercropping	2033±64	1842±78	1724±66	4.16±0.44	3.05±0.65	1.20±0.15
Maize/ <i>Sedum</i> intercropping	1918±89	1668±39	1567±12	3.45±0.58	2.38±0.33	0.37±0.01
Maize/ <i>Sedum</i> intercropping	1685±31	1496±71	1368±36	2.98±0.47	3.00±0.12	0.26±0.08
Maize/ <i>Sedum</i> intercropping	1561±106	1579±114	1202±44	3.42±0.64	2.70±0.14	0.35±0.05

Values are means ± SD.

**Table 5** Total zinc and cadmium concentrations in soils under different plant densities (mg kg<sup>-1</sup>)

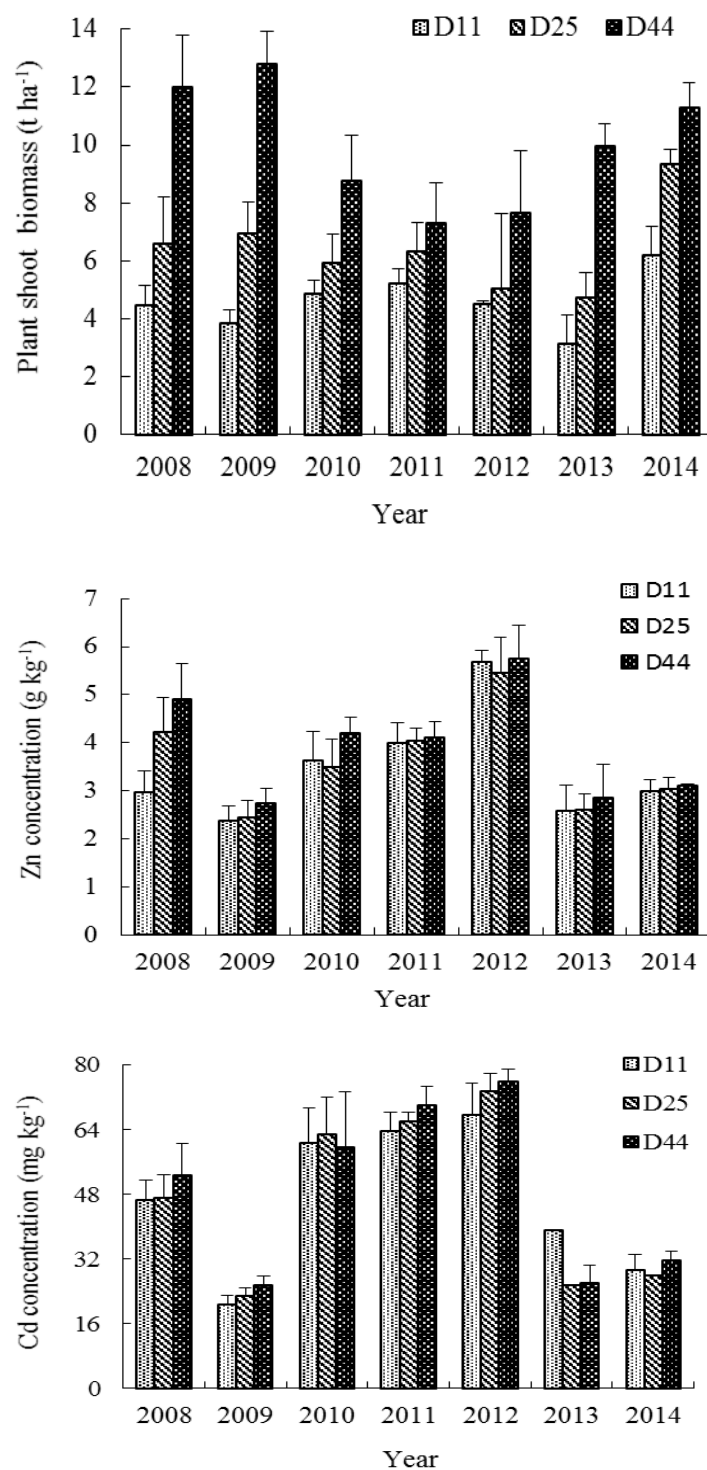
Plant	Zn				Cd			
density	2008	2009	2013	2014	2008	2009	2013	2014
CK	1312±50 a	1298±96 b	1265±72 a	1221±87 a	3.02±0.2 0a	3.11±0.3 4a	2.90±0.9 2a	3.29±0.3 7a
D11	1288±43 a	1188±11 b	1053±16 b	1037±62 b	2.94±0.3 5a	2.25±0.1 1b	1.19±0.3 6b	0.82±0.3 2b
D25	1272±67 a	1294±38 b	1044±41 b	978±41b	2.67±0.1 6a	1.96±0.0 1b	1.20±0.5 3b	0.56±0.3 1b
D44	1240±70 a	1489±13 a	1012±34 b	934±32b	2.40±0.2 2a	1.69±0.0 9b	0.94±0.2 5b	0.37±0.7 4b

Values are means ± SD; D11, D25, D44 were four planting densities with 110, 250, 440 thousand seedlings of *Sedum plumbizincicola* per hectare, respectively. Different letters in each column indicate the significant differences between treatments (p<0.05).

**Table 6** Plant metal uptake and decrease in soil metal concentrations after long-term repeated phytoextraction in two field experiments (kg ha<sup>-1</sup>)

<b>Experiment 1: Phytoextraction using intercropping</b>						
Treatment	Shoot metal uptake			Soil metal decrease		
	Maize	<i>Sedum</i>	Maize/ <i>Sedum</i>	Maize	<i>Sedum</i>	Maize/ <i>Sedum</i>
Zn	13.3±2.1	198±13	208±9	1069±45	1135±73	1238±102
Cd	0.08±0.01	5.51±0.29	4.07±0.16	3.76±0.33	5.62±0.27	6.65±1.00
<b>Experiment 2: Effect of planting density on phytoextraction efficiency</b>						
Treatment	Shoot metal uptake			Soil metal decrease		
	D11	D25	D44	D11	D25	D44
Zn	108±10	160±23	290±14	679±61	794±49	886±77
Cd	1.51±0.22	2.12±0.13	3.59±0.56	2.90±0.39	3.20±0.11	3.57±0.17

Values are means ±SD; D11, D25, D44 were four planting densities with 110, 250, 440 thousand seedlings of *Sedum plumbizincicola* per hectare, respectively.



**Figure 1** Shoot biomass and heavy metal concentrations in aboveground parts of *Sedum plumbizincicola* in the planting density field experiment Values are means  $\pm$  SD.