Phytoremediation of Heavy-Metal-Contaminated Soil in a Reclaimed Dredging Area Using *Alnus* Species

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ABSTRACT: To investigate the possible applications of plants to remediate heavy-metal-contaminated soil, a pilot experiment was performed for four years in a reclaimed dredging area using two *Alnus* species, i.e., *Alnus firma* and *Alnus hirsuta*. In a comparison of phytomass of the two species at two different planting densities, the phytomass of *Alnus* planted at low density was twice as high as that of *Alnus* planted at high density after four years. The *Alnus* species showed active acclimation to the heavy-metal-contaminated soil in a reclaimed dredging area. *A. hirsuta* showed greater accumulation of phytomass than *A. firma*, indicating that it is the better candidate for the phytoremediation of heavy-metal-contaminated soils. In the pilot system, *Alnus* plants took metals up from the soil in the following order; Pb > Zn > Cu > Cr > As > Cd. Uptake rates of heavy metals per individual phytomass was higher for *Alnus* spp. planted at low density than those planted at high density in the pilot system. Low plant density resulted in higher heavy metal uptake per plant, but the total heavy metal concentration was not different for plants planted at low and high density, suggesting that the plant density effect might not be important with regard to total uptake by plants. The quantity of leached heavy metals below ground was far in excess of that taken up by plants, indicating that an alternative measurement is required for the removal of heavy metals that have leached into ground water and deeper soil. We conclude that *Alnus* species are potential candidates for phytoremediation of heavy-metal- contaminated surface soil in a reclaimed dredging area

Key words: *Alnus* species, Density, Heavy metal, Phytoremediation, Reclaimed dredging area

INTRODUCTION

The use of heavy metals in many industrial applications has led to their wide distribution in wastewaters, sediments and soils. As a result of the high toxicity of heavy metals and their non-biodegradable nature, metal-polluted soils have thus become one of the most serious environmental problems (Bhandari et al. 2007). Phytoremediation is an emerging technology that uses certain plants to clean up soil, water, and air contaminated with environmental pollutants (Martin et al. 2007). It is clean, efficient, inexpensive and nonenvironmentally disruptive, when compared to processes that require the excavation of soil (Paraskiewicz and Dlugomski 2007). The term "phytoremediation" comes from the Greek φυτο (phyto) = plant, and the Latin "remdium" = restoring balance, or remediating (Sheehan 1997) and refers to a diverse collection of plant-based technologies that use either naturally-occurring or genetically-engineered plants to clean contaminated environments. High natural levels of metals occasionally occur in soils as a result of geological processes, but elevated metal concentrations in soil are mostly due to human industrial activities. Remediation technologies developed for metal-contaminated soils generally involve: 1) allowing heavy metals to remain at the polluted site after decreasing their availability by solidification/stabilization processes, or 2) removing heavy metals from soil by e.g., phytoremediation or soil extraction. Metal stress can cause damage and even death in sensitive plants. Most higher plants have a limited ability to cope with non-essential metals and moderately increased concentrations of essential metals. However, certain wild and crop plants, so-called hyperaccumulators, are able to accumulate large amounts of heavy metals in their aerial parts. Trees are especially

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attractive plants for revegetation of metal-polluted sites. However, most information about metal tolerance is derived from studies of a handful of herbaceous species; few studies have investigated metal tolerance in woody species and their possible utility for land reclamation (Scott 2000). Vegetation develops best when the plant-root–microorganism-soil associations are established and optimally conditioned in the rhizosphere (Khade and Adgoleya 2007). The potential of metal-tolerant and metal-accumulating plants for use in phytoremediation of contaminated soils in the study area, a reclaimed dredging area has been considered. The study area is almost flat and is dominated by dense growth of *Phragmites communis*, interspersed with *Alnus firma* and *A. hirsuta* in a mosaic pattern. *Alnus* species have the capability of fixing nitrogen and utilizing insoluble metals using inoculated mycorrhiza (Becerra et al. 2005). To examine the effect of planting density on soil heavy metal uptake by plants and runoff from the planting basin, and to evaluate the potential use of *Alnus* species as a phytoremediator to reduce heavy metals contamination, we planted *Alnus* species at two different densities in a reclaimed dredging area and monitored the results for four years.

MATERIALS AND METHODS

Materials used for the analyses of vegetation and soil were obtained from a reclaimed dredging area in Gwangyang Bay, South Korea, which we described in detail in a previous paper (Lee et al. 2009). There was no additional physical disturbance to the Gwangyang Bay reclaimed dredging area for 15 to 20 years after reclamation. The soil in the area was categorized as sandy soil (Lee et al. 2009). From April 2004 to September 2007, we conducted field experiments in two permanent plots planted with two landscape tree species at two planting densities (Fig. 1). The size of the combined experimental plot was 33 m (W) x 50 m (L) x 1 m (H). In the pilot planting plot, 100 individuals of each tree species were planted in high-density quadrats (10 m x 10 m) and 49 individuals of each species of the same age and size were planted at low density in 10 m x 20 m quadrats. Saplings of *A. firma* and *A. hirsuta* planted in the experimental plots averaged 100.2 cm and 100.3 cm in height, respectively. After planting of the saplings, the pilot planting system was irrigated with a watering cart weekly. Plants (*A. firma* and *A. hirsuta*) collected from this area were analyzed for their heavy metal contents (As, Cr, Pb, Cu, Cd, and Zn) to evaluate their phytoremediation capability. After 4 years, to estimate phytomass and production for each plant, we sequentially harvested plant parts during the course of the

growing season (Whittaker and Marks 1975). The trees of each species were separated into their reproductive parts, leaves, branches, stems, main roots, coarse roots (2-8 mm), and fine roots (<2 mm). Subsamples of these materials were dried at 80°C until their mass remained constant and their masses were then recorded. Digestion was then carried out in a microwave digester (Mars-Xpress, CEM, USA). The digested samples were diluted to 15 mL in a volumetric plastic tube with double distilled water. The heavy metal concentrations of soil samples and plant tissues collected soon after planting and after four years were determined using an inductively coupled plasma mass spectrometer (ICP-Ms) (ELAN 6100, Perkin-Elmer, USA) at the Reliability Assessment Center, Research Institute of Industrial Science & Technology (RIST). The accuracy of the microwave digester system was tested with Certified Reference Material (CRM): Peach leaves (SRM 1547) and light sandy soil (CRM 142R) from the National Institute of Standard & Technology (NIST).
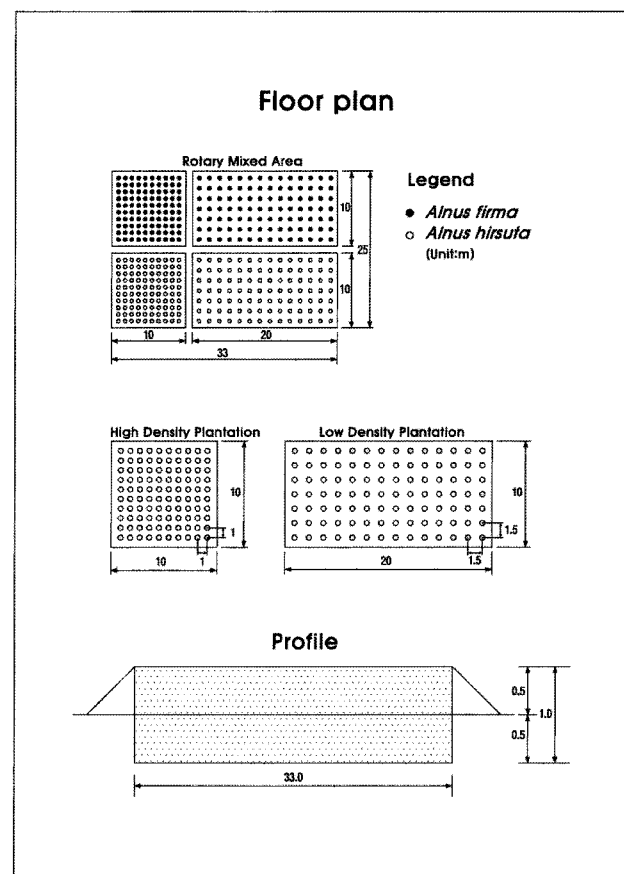

Fig. 1. Layout of the planting condition of phytoremediation pilot system.

Table 1. Heavy metal concentrations in soil samples (n = 20 samples) from the phytoremediation pilot system

Division		Heavy metal concentrations (mg/kg)					
		As	Cr	Pb	Cu	Cd	Zn
Study sites	Min.	0.139	48.6	42.6	11.7	0.78	266
	Max.	0.349	141.9	482.0	57.9	4.02	2,230
	Mean ± STDEV	0.236 ± 0.078	75.4 ± 23.7	267.4 ± 99.1	27.8 ± 10.4	1.88 ± 0.79	960 ± 556
Criterion of apprehension at industry area		20	12 (Cr^{6+})	400	200	12	800

Table 2. Heavy metal concentrations of reference plants (n = 12 samples) used on the phytoremediation pilot system early in the phytoremediation process

Species	Organs	Phytomass (g.d.w./indiv.)	Heavy metal concentrations (mg/kg)					
			As	Cr	Pb	Cu	Cd	Zn
Alnus firma	Leaves	25.0 ± 2.3	0.13 ± 0.04	0.33 ± 0.11	1.45 ± 0.59	1.77 ± 0.29	0.04 ± 0.04	13.65 ± 3.06
	Branches & Stem	24.8 ± 4.9	0.23 ± 0.12	0.27 ± 0.05	2.95 ± 0.54	2.91 ± 0.54	0.28 ± 0.06	11.47 ± 1.15
	Root	13.0 ± 2.6	0.06 ± 0.01	0.32 ± 0.03	5.82 ± 0.30	2.91 ± 0.30	0.20 ± 0.02	10.44 ± 1.03
	Total (µg/indiv.)	62.8 ± 3.7	**10.00 ± 5.11**	**19.38 ± 3.93**	**183.54 ± 3.98**	**156.14 ± 28.85**	**10.44 ± 1.65**	**764.37 ± 98.78**
Alnus hirsuta	Leaves	32.9 ± 3.6	0.11 ± 0.02	0.34 ± 0.02	1.57 ± 0.28	2.70 ± 0.47	0.12 ± 0.02	10.95 ± 1.19
	Branches & Stem	31.8 ± 1.5	0.18 ± 0.03	0.28 ± 0.03	4.90 ± 1.26	4.44 ± 1.03	0.27 ± 0.08	11.17 ± 1.35
	Root	20.3 ± 2.0	0.11 ± 0.03	0.58 ± 0.05	8.32 ± 0.87	6.60 ± 1.52	0.46 ± 0.14	11.51 ± 2.44
	Total (µg/indiv.)	84.9 ± 7.0	**11.96 ± 2.34**	**31.91 ± 4.97**	**378.56 ± 90.30**	**363.71 ± 41.10**	**22.04 ± 6.15**	**951.25 ± 133.36**

RESULTS AND DISCUSSION

Soil heavy metal concentrations early in the experiment

Soil heavy metal concentrations early in the phytoremediation pilot system are presented in Table 1. Concentrations of heavy metals in the study area were 0.14-0.35 mg/kg for As, 48.60-141.90 mg/kg for Cr, 42.6-482.0 for Pb, 11.7-57.9 for Cu, 0.78-4.02 for Cd, and 266-2,230 mg/kg for Zn (Table 1). When planting for the pilot system experiment started, levels of three heavy metals, Cr, Pb and Zn, were over the baseline levels delineated by the Soil Environment Apprehension Guideline of Korea (Ministry of Environment 2007). Generally, concentrations of heavy metals in normal soils range from 0.1 to 102 mg/ kg for As, 0.005 to 3,950 mg/kg for Cr, 1 to 6,900 mg/ kg for Pb, 0.03 to 550 mg/kg for Cu, 0.1 to 345 mg/kg for Cd, and 0.15 to 5,000 mg/kg for Zn (Eapen et al. 2007). Concentrations of this study soil were lower than those in previous studies due to the heterogeneous mixture in the planting basin of reclaimed dredging soil and fresh mountain soil with low organic matter (<1%), and electric conductivity (<0.5 µS/cm) (Lee et al. 2009). The binding mechanisms of heavy metals are complex and vary with the composition of soil, soil acidity and redox condition (Thangavel and Subburaam 2004). The bioavailability and mobility of heavy metals in soils is dependent upon the redistribution processes between solution and solid phases and among solid phase components. The rates of redistribution of metals and their binding intensity in soils are affected by the metal species, loading levels, ageing and soil properties (Han et al. 2003).

Heavy metals concentrations of reference plant organs

Heavy metal concentrations in the reference plants used in the phytoremediation pilot system are shown in Table 2. The low concentrations of As, Cr, Pb, Cu, Cd and Zn per individual in the *A. firma* and *A. hirsuta* saplings were good reference criteria, and were within the lower limits for typical background concentrations in higher plants (Bowen 1979).

Phytomass after 4 years

Plant phytomass per individual in the pilot system after four years is shown in Table 3. Total phytomass of *A. firma* increased 95-fold from 63 g·d.w./indiv. to 5,967 g·d. w./indiv. in the high-density quadrat and 197-fold from 63 g·d.w./indiv. to 12,397 g·d.w./indiv. in the low-density quadrat. After four years, total phytomass for *A. hirsuta* increased 125-fold from 85 g·d.w./indiv. to 10,659 g·d. w./indiv. in the high-density quadrat, and 222-fold from 85 g·d.w./indiv. to 18,908 g·d.w./indiv. in the low-density quadrat. Phytomass of the two species was twice as high in the low-density quadrats as it was at high density after four years (Table 3). After four years, the relative growth rates of *A. firma* and *A. hirsuta* were 1,476 and 2,643 g·d.

Table 3. Components of the phytomass (g. DW./ indiv. n = 12 samples) of *Alnus firma* and *A. hirsuta* in the phytoremediation pilot system under high-density and low-density planting conditions

Plant parts	Alnus firma		Alnus hirsuta	
	High density	Low density	High density	Low density
Reproductives*	311 ± 30	813 ± 168	160 ± 14	210 ± 10
Leaves*	786 ± 15	1,601 ± 25	1,241 ± 72	2,691 ± 37
Branches*	921 ± 131	1,927 ± 257	1,731 ± 97	4,931 ± 52
Stem*	2,818 ± 272	5,516 ± 191	5,620 ± 203	7,700 ± 63
Main root*	680 ± 143	1,716 ± 145	1,221 ± 69	2,280 ± 44
Coarse roots*	295 ± 51	542 ± 72	502 ± 55	819 ± 15
Fine roots*	156 ± 22	282 ± 5	184 ± 17	277 ± 5
Total	5,967 ± 664	12,196 ± 825	10,659 ± 527	19,187 ± 704

*: Significant at the 0.05 level by t-test

Table 4. Heavy metal concentrations (n = 12 samples) of *Alnus firma* and *A. hirsuta* in mg per kg dry weight of plant parts on the phytoremediation pilot system

Species	Plant parts	Heavy metal concentrations (mg/kg)											
		As		Cr		Pb		Cu		Cd		Zn	
		High	Low	High	Low	High	Low	High	Low	High	Low	High	Low
Alnus firma	Reproductives	0.23 ±0.02	0.13 ±0.01	0.51 ±0.01	0.77 ±0.09	43.8 ±3.3	42.4 ±2.6	8.5 ±0.8	8.0 ±0.2	0.05 ±0.01	0.04 ±0.03	33.9 ±3.9	31.4 ±1.0
	Leaves	0.11 ±0.01	0.18 ±0.01	0.88 ±0.01	0.97 ±0.11	70.5 ±3.7	81.4 ±1.0	7.1 ±0.4	6.9 ±0.4	0.05 ±0.01	0.06 ±0.01	34.9 ±3.5	44.2 ±3.0
	Branches	0.14 ±0.03	0.11 ±0.01	0.78 ±0.01	0.51 ±0.05	53.0 ±2.9	57.1 ±1.0	6.6 ±0.5	5.1 ±0.5	0.13 ±0.01	0.18 ±0.02	31.7 ±3.5	28.4 ±0.9
	Stem	0.16 ±0.01	0.14 ±0.01	0.42 ±0.01	0.53 ±0.02	49.0 ±1.3	38.1 ±9.7	3.6 ±0.4	2.8 ±0.4	0.04 ±0.01	0.04 ±0.01	6.4 ±2.9	11.5 ±1.3
	Main root	0.19 ±0.01	0.11 ±0.01	1.07 ±0.09	1.20 ±0.03	52.6 ±2.4	44.2 ±3.1	5.3 ±0.2	3.7 ±0.1	0.07 ±0.01	0.10 ±0.01	10.9 ±0.5	13.7 ±1.1
	Coarse roots	0.12 ±0.01	0.41* ±0.01	0.81 ±0.05	1.34* ±0.09	44.7 ±1.6	52.7 ±2.0	4.4 ±0.5	4.3 ±0.3	0.06 ±0.01	0.07 ±0.02	17.1 ±0.5	12.1 ±0.6
	Fine roots	0.41 ±0.01	0.23 ±0.01	1.51 ±0.03	1.83 ±0.13	81.4 ±3.3	55.5* ±4.9	7.0 ±0.2	8.3 ±0.4	0.17 ±0.01	0.21 ±0.02	36.9 ±3.3	35.4 ±0.8
Alnus hirsuta	Reproductives	0.13 ±0.01	0.13 ±0.01	0.29 ±0.01	0.37 ±0.04	85.6 ±5.0	80.2 ±2.1	17.6 ±1.8	10.5 ±0.3	0.03 ±0.01	0.05 ±0.01	40.7 ±9.5	50.7 ±2.3
	Leaves	0.07 ±0.05	0.11 ±0.01	0.82 ±0.01	1.07 ±0.16	84.6 ±3.6	81.4 ±1.6	7.8 ±0.9	6.3 ±0.2	0.05 ±0.01	0.04 ±0.01	42.0 ±10.2	32.5 ±0.8
	Branches	0.20 ±0.01	0.20 ±0.01	0.35 ±0.01	0.50 ±0.02	54.1 ±3.4	42.7 ±2.7	4.9 ±0.2	3.6 ±0.3	0.14 ±0.01	0.11 ±0.01	29.1 ±1.3	33.1 ±2.2
	Stem	0.13 ±0.01	0.17 ±0.05	0.45 ±0.01	0.62 ±0.02	47.1 ±2.0	43.0 ±2.5	2.6 ±0.2	2.4 ±0.1	0.06 ±0.01	0.05 ±0.01	10.0 ±0.3	11.1 ±1.1
	Main root	0.25 ±0.01	0.29 ±0.03	0.85 ±0.01	0.62 ±0.01	46.3 ±2.1	42.8 ±2.9	3.4 ±0.2	2.6 ±0.2	0.07 ±0.01	0.09 ±0.01	14.7 ±0.9	16.5 ±0.7
	Coarse roots	0.20 ±0.01	0.21 ±0.02	1.11 ±0.01	1.13 ±0.01	54.8 ±1.8	48.8 ±3.8	4.0 ±0.2	3.4 ±0.3	0.08 ±0.01	0.06 ±0.01	10.9 ±0.6	11.3 ±1.1
	Fine roots	0.05 ±0.01	0.04 ±0.01	2.30 ±0.02	2.86 ±0.06	59.1 ±3.2	51.0 ±3.0	8.3 ±0.4	7.6 ±0.6	0.23 ±0.01	0.18 ±0.01	34.6 ±4.1	33.5 ±1.6

* : Significant at the 0.05 level by t-test

w./indiv./yr. at high density and 3,033 and 4,775 g·d.w./indiv./yr. at low density. Yield of phytomass per individual for the *Alnus* species in the high-density quadrates was greater than that in the low-density quadrates. However, the growth per unit area did not differ with planting density. This suggests that *Alnus* species adapted very well to the reclaimed dredging area and has the capability to grow quickly, fix nitrogen, and utilize insoluble nutrients via inoculated mycorrhiza. Alder roots are associated with ectomycorrhizal, arbuscular mycorrhizal, and actinorrhizal
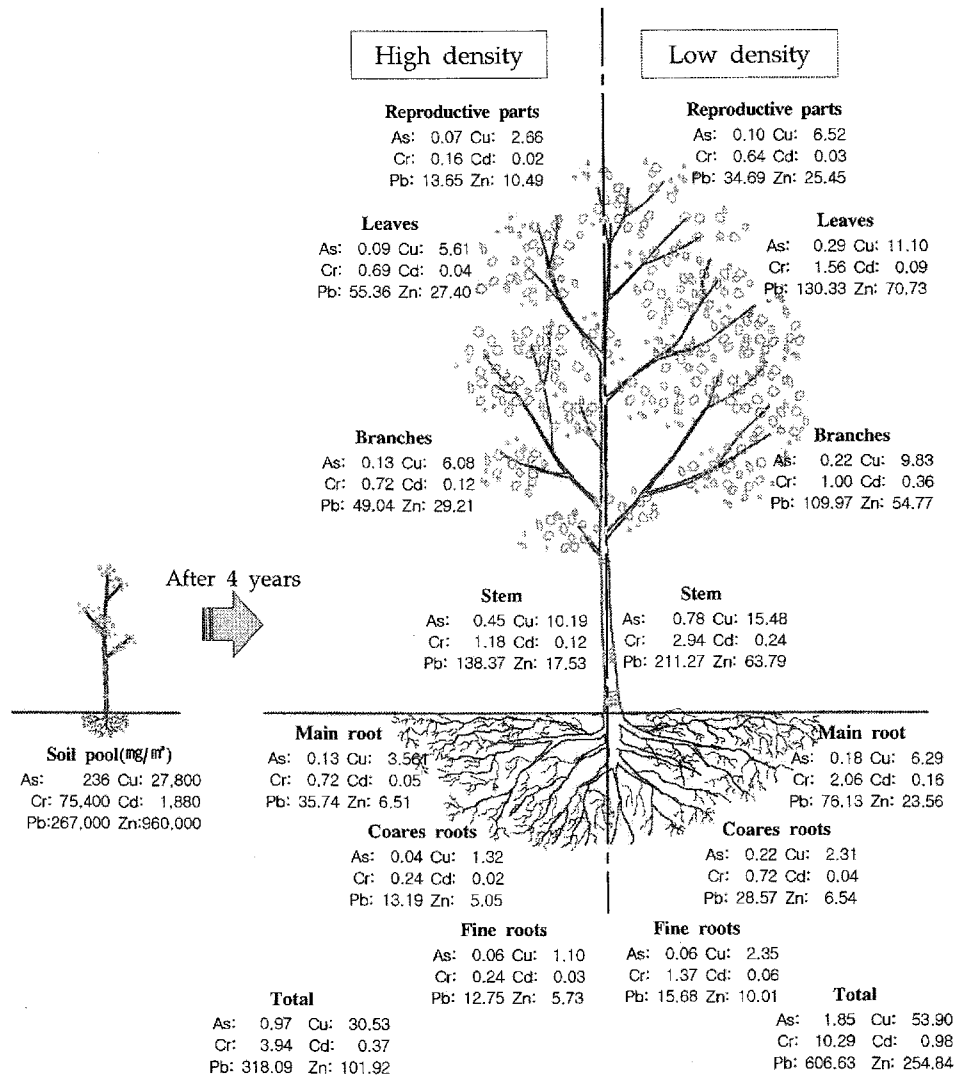
Fig. 2. Uptake concentration of heavy metals in mg per parts of individual of *Alnus firma* according to the planting condition on the phytoremediation pilot system.

symbionts (Zhuang et al. 2007, Lee et al. 2009). All of these symbionts are known to be beneficial to the host, contributing to a better nutritional status and pathogen defense and thus enhancing the capacity for establishment of individual plants (Becerra et al. 2005).

Heavy metal concentrations of plant organs after 4 years

Heavy metal concentrations in *A. firma* and *A. hirsuta* plant parts in the phytoremediation pilot system are shown in Table 4.

Heavy metal concentrations of most plant parts in the phytoremediation pilot system did not differ between the two different planting densities except for coarse and fine roots of *A. firma*, which differed significantly in their concentrations of some heavy metals at different planting densities (Table 4). Typical background As, Cr, Pb, Cu, Cd, and Zn concentrations of higher plants are 0.2-7, 0.03-

10, 1-13, 5-15, 0.1-2.4, 20-400 mg/kg·d.w, respectively (Bowen 1979). Critical limits for Cr, Pb, Cu, Cd, and Zn of higher plants were 1-2, 10-20, 15-20, 5-10, and 150-200 mg/kg·d.w., respectively (Sauerbeck 1982). Compared to Sauerbeck's critical limit values, As, Cr, Cd, and Zn showed higher values, Pb showed lower value, and Cu exhibited reported critical limit value range. The transfer of As from soil to plant is low for most plant species for several reasons: (1) low bioavailability of As in soil, (2) restricted uptake by plant roots, (3) limited translocation of As from roots to shoot, and (4) As phytotoxicity at relatively low concentrations in plant tissues (Willey 2007). However, *Pteris vittata* accumulated as much as 27,000 mg-As/kg·d.w. in fronds growing on an arsenate site (Wang et al. 2002). Hyperaccumulators are defined as plants that can accumulate 100 m/kg Cd, 1000 mg/kg As, Cu, Cr, Pb, or 10,000 mg/kg Zn (Baker and Brooks 1989). McCutcheon
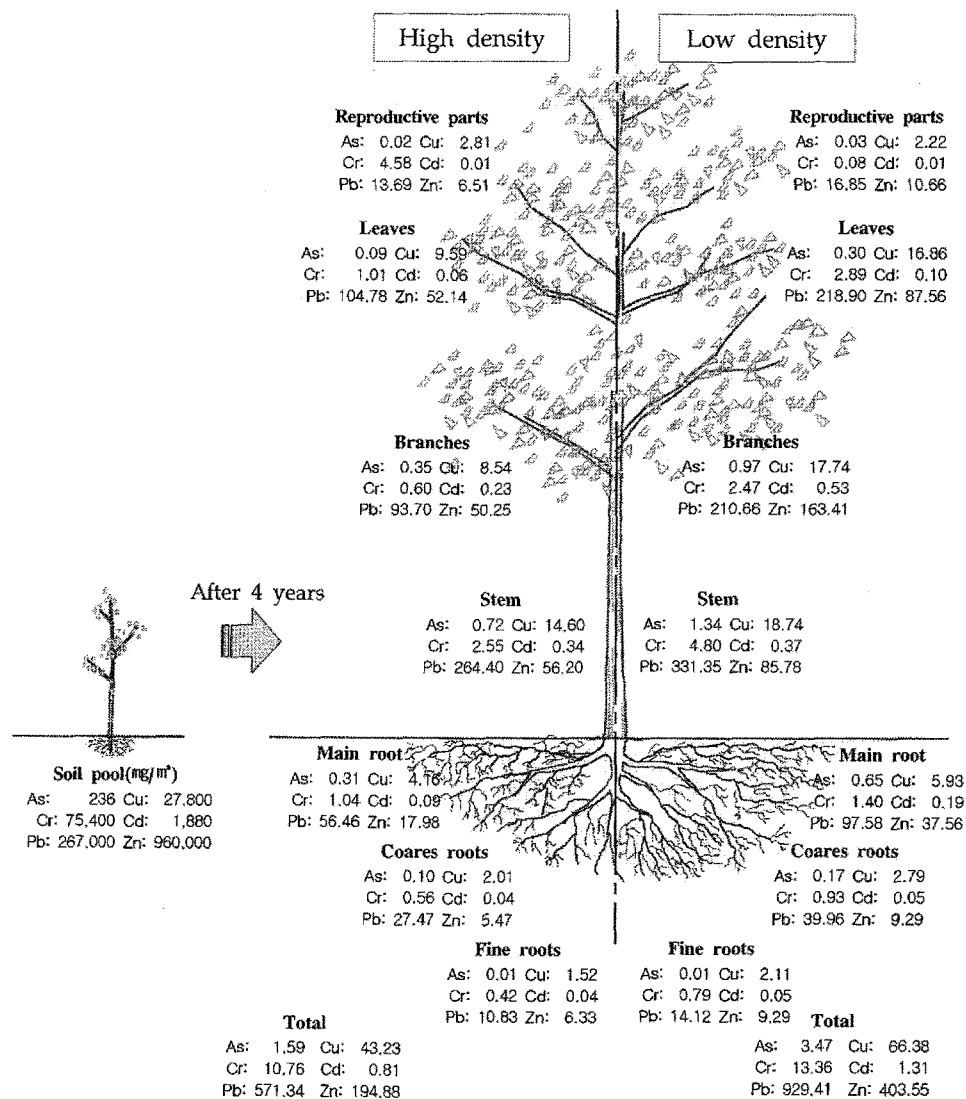
High density   Low density

After 4 years

Reproductive parts
As: 0.02 Cu: 2.81
Cr: 4.58 Cd: 0.01
Pb: 13.69 Zn: 6.51

Reproductive parts
As: 0.03 Cu: 2.22
Cr: 0.08 Cd: 0.01
Pb: 16.85 Zn: 10.66

Leaves
As: 0.09 Cu: 9.59
Cr: 1.01 Cd: 0.06
Pb: 104.78 Zn: 52.14

Leaves
As: 0.30 Cu: 16.86
Cr: 2.89 Cd: 0.10
Pb: 218.90 Zn: 87.56

Branches
As: 0.35 Cu: 8.54
Cr: 0.60 Cd: 0.23
Pb: 93.70 Zn: 50.25

Branches
As: 0.97 Cu: 17.74
Cr: 2.47 Cd: 0.53
Pb: 210.66 Zn: 163.41

Stem
As: 0.72 Cu: 14.60
Cr: 2.55 Cd: 0.34
Pb: 264.40 Zn: 56.20

Stem
As: 1.34 Cu: 18.74
Cr: 4.80 Cd: 0.37
Pb: 331.35 Zn: 85.78

Soil pool(mg/m²)
As: 236 Cu: 27,800
Cr: 75,400 Cd: 1,880
Pb: 267,000 Zn: 960,000

Main root
As: 0.31 Cu: 4.76
Cr: 1.04 Cd: 0.09
Pb: 56.46 Zn: 17.98

Main root
As: 0.65 Cu: 5.93
Cr: 1.40 Cd: 0.19
Pb: 97.58 Zn: 37.56

Coares roots
As: 0.10 Cu: 2.01
Cr: 0.56 Cd: 0.04
Pb: 27.47 Zn: 5.47

Coares roots
As: 0.17 Cu: 2.79
Cr: 0.93 Cd: 0.05
Pb: 39.96 Zn: 9.29

Fine roots
As: 0.01 Cu: 1.52
Cr: 0.42 Cd: 0.04
Pb: 10.83 Zn: 6.33

Fine roots
As: 0.01 Cu: 2.11
Cr: 0.79 Cd: 0.05
Pb: 14.12 Zn: 9.29

Total
As: 1.59 Cu: 43.23
Cr: 10.76 Cd: 0.81
Pb: 571.34 Zn: 194.88

Total
As: 3.47 Cu: 66.38
Cr: 13.36 Cd: 1.31
Pb: 929.41 Zn: 403.55

Fig. 3. Uptake concentration of heavy metals in mg per parts of individual of *Alnus hirsuta* according to the planting condition on the phytoremediation pilot system.

et al. (2003) reported that *Dicoma niccolifera* accumulated as much as 1,000 mg- Cr/kg·d.w., *Salvinia molesta* accumulated 200 mg-Pb/kg·d.w, and *Thlaspi caerulescens* accumulated 2,130 mg-Cd/kg and 10,000 mg-Zn/kg·d.w.

Heavy metal concentrations of individual plants after four years

Heavy metal concentrations of individual of *Alnus* species after four years of growth in the phytoremediation pilot system are shown in Figs 2 and 3. In high-density quadrats, uptakes of As, Cr, Pb, Cu, Cd, and Zn per individual were 1.0, 3.9, 318.1, 30.5, 0.4, and 101.9 mg/indv. for *A. firma* and 1.6, 10.8, 571.3, 43.2, 0.8, and 194.9 mg/indiv. for *A. hirsuta*. Heavy metal uptakes in the low-density quadrats were 1.9, 10.3, 606.6, 53.9, 1.0, and 254.8 mg/indiv. for *A. firma* and 3.5, 13.4, 929.4, 66.4, 1.3, and 403.6 mg/indiv. for *A. hirsuta* (Figs 2 and 3). Heavy metal concentrations per individual for *A. firma* and *A. hirsuta* were higher at low density than at high density. The bulk of metal in the soil is commonly found as insoluble compounds unavailable for transport into roots from the aqueous phase. Binding and immobilization of the toxic metals within the soil matrix can significantly restrict their uptake and removal from the site. The bioavailability of the metals and other toxic substances, however, can be enhanced by manipulating the rhizosphere of the potential remediator plants by changing soil pH (lowering of pH is recommended to increase the bioavailability of heavy metals), adding chelating agents, using appropriate fertilizers (ammonium containing fertilizers), altering soil ion composition, adding adequate consortia of soil microbes and phytosiderophores and soil exudate

Table 5. Secular changes in heavy metal concentrations (mg/kg, n = 12 samples), percentage of uptake by plants and runoff from the soil in the phytoremediation pilot system

Species	Heavy Metals	As		Cr		Pb		Cu		Cd		Zn	
		High	Low	High	Low	High	Low	High	Low	High	Low	High	Low
Alnus firma	Orig. conc. in soil	0.236		74.5		267.0		27.8		1.88		960	
	Conc. in soil after 4 years	0.188	0.190	33.3	43.3	213	204	17.6	18.3	1.20	1.33	764	762
	Uptake (%)	0.56	0.79	0.01	0.01	0.12	0.22	0.10	0.19	0.02	0.05	0.01	0.03
	Runoff (%)	20.3	19.5	55.9	42.6	20.2	23.6	36.7	34.2	36.2	29.2	11.2	11.4
Alnus hirsuta	Orig. conc. in soil	0.236		74.5		267.0		27.8		1.88		960	
	Conc. in soil after 4 years	0.170	0.182	46.0	48.9	238	203	19.3	18.0	1.11	1.23	777	760
	Uptake (%)	0.67	1.47	0.01	0.02	0.21	0.35	0.16	0.24	0.05	0.07	0.02	0.05
	Runoff (%)	30.0	22.9	39.0	35.2	10.9	24.0	30.6	35.3	40.9	34.6	9.7	11.6

Table 6. Uptake of heavy metals by *Alnus* species (n = 12) according to planting condition on the phytoremediation pilot system after four years

(Unit : g/ha)

Species	Heavy metals	As		Cr*		Pb*		Cu*		Cd*		Zn	
		High	Low	High	Low	High	Low	High	Low	High	Low	High	Low
Alnus firma		9.7 ±0.5	9.1 ±1.4	39.4 ±2.7	50.4 ±7.0	3,180 ±271	2,972 ±697	305.2 ±30.8	264.0 ±59.0	3.7 ±0.3	4.7 ±1.2	1,019 ±65	1,248 ±154
Alnus hirsuta		15.9 ±0.8	17.0 ±5.7	107.6 ±7.9	65.4 ±8.1	5,713 ±256	4,554 ±350	432.3 ±15.2	325.2 ±12.6	8.1 ±3.1	6.4 ±0.9	1,948 ±166	1,977 ±250

*: Significant at the 0.05 level by t-test

management (Singh and Tripathi 2007). A key to effective phytoremediation, especially phytoextraction, is to enhance metal phyto-availability and to sustain adequate metal concentrations in the soil solution for plant uptake (Lombs et al. 2001).

Heavy metal concentrations in soil after four years

Heavy metal concentrations in the soil at the phytoremediation pilot system after four years are shown in Table 5. As, Cr, Pb, Cu, Cd, and Zn concentrations in the soil of the phytoremediation pilot system after four years were 0.19, 37.91, 239.19, 24.62, 1.25 and 793.05 mg/kg on average in the high-density quadrats and 0.19, 39.45, 228.02, 20.27, 1.30 and 785.70 mg/kg on average in the low-density quadrats. There was no difference in heavy metal concentrations in soil or runoff between the high-density and low-density quadrats. The rate of decrease for heavy metal concentrations in the soil of the pilot system after four years of phytoremediation using *A. firma* and *A. hirsuta* were 20.34% and 27.96% for As, 55.70% and 39.03% for Cr, 20.22% and 10.49% for Pb, 36.69% and 30.6% of Cu, 35.64% and 35.11% for Cd, and 20.42% and 19.06% for Zn in high-density quadrats, and 19.49% and 22.88% for As, 42.61% and 35.19% for Cr, 23.60% and

23.97% for Pb, 34.17% and 35.25% for Cu, and 28.72% and 34.04% for Cd, and 20.52% and 20.83% for Zn in low-density quadrats (Table 5). After four years, heavy metal concentrations in the soil had been reduced more by runoff from rainfall than uptake by plants, which probably resulted from the soil characteristics in the reclaimed dredging area. Sandy soil comprises the major component of the soil in the study area, and during the application of soil after dredging, reclaimed dredging soil was mixed with an equal quantity of fresh soil, resulting in an increase in soil porosity that enhanced the rainfall wash-out effect (Lee et al. 2009). Further remediation is needed to address the problem of heavy metals in the ground water at the study site.

Uptake of heavy metals by *Alnus* species

Uptakes of heavy metals from the soil by *Alnus* species per unit area in the phytoremediation pilot system after 4 years are shown in Table 6. Uptakes of As, Cr, Pb, Cu, Cd and Zn in the high-density quadrats were 9.73, 39.43, 3,180.91, 305.28, 3.75 and 1,019.22 g/ha for *A. firma* and 15.94, 107.60, 5,713.40, 432.32, 8.14 and 1,948.81 g/ha for *A. hirsuta*. And uptakes in the low-density quadrats were 9.07, 50.40, 2,972.50, 264.09, 4.79 and 1,248.73 g/

ha for *A. firma* and were 17.02, 65.46, 4,554.12, 325.26, 6.41 and 1,977.39 g/ha for *A. hirsuta*. Uptake of As and Zn by the two species was very similar under different density planting conditions, uptake of Cr, Pb and Cu was higher in the high-density quadrats than the low-density quadrats and uptake of Cd was higher at low density for *A. firma* and was higher at high density for *A. hirsuta*. Major obstacles to successful phytoremediation must be overcome before phytoremediation can be promoted as an effective solution for heavy-metal-contaminated soil: yield and metal uptake rates have to be increased dramatically in order to allow remediation within reasonable periods of time (Han et al. 2003, Lee at al. 2009). Plant selection for phytoremediation generally focuses on targeting species that accumulate or metabolize toxic compounds in an organ that is easy to remove from the area (Lomb et al. 2001). Generally, fast-growing plants with high phytomass and different kinds of root systems suitable for cleanup of pollutants at different depths are considered as ideal phytoremediators (Bhandari et al. 2007). However, the plants must also be tolerant enough of the target toxicants to survive and grow vigorously in contaminated sites and should be suitable for the agro-climatic conditions of the area under cleanup (Singh and Tripathi 2007). Selection of appropriate plants for phytoremediation should be considered during every step of the developmental process. Our results suggest that plant selection and density of planting were key factors affecting the success of heavy metal phytoremediation approaches.

CONCLUSIONS

We performed a phytoremediation experiment with two *Alnus* species planted at different densities in a reclaimed dredging area for four years. Total phytomass per plant was twice as high in the low-density than the high-density quadrats, suggesting that trees at low density had an advantage in resource utilization over trees at high density. Heavy metal uptake and runoff from the planting basin under high and low planting density showed irregular patterns because of the heterogeneous distribution of heavy metals in soil and different soil's physical trait and rain-wash out effect. Heavy metal uptake per individual was higher at low density than at high density, but total heavy metal concentrations in the plants per unit area was not different between the low- and high-density quadrats, suggesting that the plant density effects might not be important in terms of total uptake by plants in a young plantation. In particular, the quantities of leached heavy metals below ground were far greater than those taken up by plants from the reclaimed dredging soil, indicating that additional measures will be required for the removal of leached heavy metals from the ground water and accumulated heavy metals in deeper soils. *Alnus* species showed active acclimation to heavy-metal-contaminated soil in the reclaimed dredging area and was capable of growing fast, fixing nitrogen, and utilizing insoluble nutrients. *A. hirsuta* showed greater phytomass and heavy metal accumulation than *A. firma*, indicating that it is the better candidate for phytoremediation of heavy-metal-contaminated soils. Additional research is required to determine the effectiveness of phytoremediation using other landscape tree species in the reclaimed dredging area.

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