



Development of a technology for commercial phytoextraction of nickel: economic and technical considerations

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Key words: hyperaccumulator, nickel, phytoextraction, phytomining, phytoremediation

Abstract

In recent R&D work, we have made progress in developing a commercial technology using hyperaccumulator plant species to phytoextract nickel (Ni) from contaminated and/or Ni-rich soils. An on-going program is being carried out to develop a genetically improved phytoextraction plant that combines favorable agronomic and Ni accumulation characteristics. Genetically diverse Ni hyperaccumulator species and ecotypes of *Alyssum* were collected and then evaluated in both greenhouse and field using serpentine and Ni-refinery contaminated soils. Large genetic variation was found in those studies. Mean shoot Ni concentrations in field-grown plants ranged from 4200 to 20 400 mg kg⁻¹. We have been studying several soil management practices that may affect the efficiency of Ni phytoextraction. Soil pH is an important factor affecting absorption of metals by plants. An unexpected result of both greenhouse and field experiments was that Ni uptake by two *Alyssum* species was reduced at lower soil pH and increased at higher soil pH. At higher pH, plant yield was improved also. In soil fertility management studies, we found that N application significantly increased plant biomass, but did not affect plant shoot Ni concentration. These findings indicate that soil management will be important for commercial phytoextraction. A number of field trials have been carried out to study planting methods, population density, weed control practices, harvest schedule and methods, pollination control, and seed processing. Such crop management studies have improved phytoextraction efficiency and provide a tool for farmers to conduct commercial production. We have done some work to develop efficient and cost-effective methods of Ni recovery. Recovery of energy by biomass burning or pyrolysis could help make phytoextraction more cost-effective. The progress made in our recent studies will enable us to apply this technology commercially in the near future.

Introduction

Development of a commercially viable technology for using hyperaccumulator plant species to phytoextract Ni from contaminated and/or Ni-rich soils requires (1) identifying or creating an ideal phytoextraction plant, (2) optimizing soil and crop management practices, and (3) developing methods for biomass processing and Ni extraction. In this paper, we report on recent progress made by our group in the development of Ni phytomining technology.

Phytoextraction employs plants to transport and accumulate high quantities of metals from soil into the harvestable parts of roots and aboveground shoots (Chaney, 1983; Chaney et al., 1997). Metal concentrations achieved by naturally occurring hyperaccumulating plant species can be 100–1000 times those that occur in non-accumulator plants growing in the same substrates. Ni hyperaccumulation has been defined as the accumulation of at least 1000 mg kg⁻¹ Ni in the dry biomass of plants grown on a natural substrate (Brooks et al., 1977; Jaffré et al., 1976). Phytoextraction can be applied either to metal-contaminated soils or to ores that cannot be economically enriched by traditional mining technology (Chaney et al., 1998). In

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the former case, phytoextraction is a type of phytoremediation, while the term 'phytomining' has been applied to the latter case, in which the economic value of the recovered metal is the primary motive. Of course, the value of the phytoextracted metal, if it is recycled, can help to defray the costs of phytoremediation.

To date, the majority of phytoremediation work has focused on cadmium, lead, and zinc (Huang and Cunningham, 1996; Li and Chaney, 1998; McGrath et al., 1993). However, Ni contamination is a problem in many soils where sewage sludge has been used as a soil amendment (McGrath and Smith, 1990). Ni concentrations as high as 385 mg kg⁻¹ have been measured in UK agricultural soils receiving sewage sludge. Background levels of Ni in the UK are around 25 mg kg⁻¹, and the European Community has set 75 mg kg⁻¹ as the maximum allowable in pasture soils (McGrath, 1995). In an extensive survey of heavy metal concentrations in US agricultural soils, Holmgren et al. (1993) reported a maximum Ni concentration of 390 mg kg⁻¹ and a mean Ni concentration of 25 mg kg⁻¹. Emissions from Ni refineries and smelters have also increased Ni (and Co) concentrations in nearby soils. For example, downwind from a Ni refinery in Port Colborne, Ontario, Canada, the concentration of Ni in the 0–5 cm layer of untilled muck soil ranged from 800 to over 6000 mg kg⁻¹ (Frank et al., 1982). Vegetable crops grown on this soil, which had an acidic pH, showed visual symptoms of Ni phytotoxicity, and yields of several Ni-sensitive vegetable crops were substantially reduced, apparently due to the presence of the soil contaminants (Frank et al., 1982). In cases such as these, remediation may be necessary to meet regulatory standards. Phytoextraction of Ni by hyperaccumulator plants is a potential method of remediating these soils (Chaney et al., 1998; Li et al., 2000).

Commercial mining of Ni is usually conducted with ores that have a high concentration of the target metal. For mining to be economically viable, ore bodies with a Ni content of at least 30 g kg⁻¹ are required. Few ore bodies of this nature occur on the earth's surface, and some of these are becoming exhausted. There are much larger areas of low-grade ore, which, however, are not economical to exploit using current mining technology. Most of these ore bodies are associated with ultramafic deposits. Weathering of the ultramafic rocks has produced what are termed serpentine soils, which are characterized by a pH of 6–8, low ratios of Ca/Mg, low levels of N, P and K, and potentially phytotoxic amounts of Ni.

The severely deficient levels of Ca, P, and N in most serpentine soils make them inhospitable to normal terrestrial plants (Nicks and Chambers, 1995, 1998). Serpentine areas are scattered throughout the world and usually support a characteristic flora. In California and Oregon, there are over 400 000 ha of serpentine soils (Alexander, 1994). In these soils, Ni is usually present at concentrations between 1.0 and 7.0 g kg⁻¹, well below the minimum Ni concentration required for modern mining technologies, but adequate to supply Ni hyperaccumulators. Thus, there is an opportunity to commercially phytomine such soils to produce biomass ash much richer in Ni than common Ni ores (Baker and Brooks, 1989; Brooks et al., 1998; Chaney et al., 1998; Nicks and Chamber, 1995; Reeves, 1992).

Chaney (1983) proposed the concept of using plant hyperaccumulators for phytoextraction. In 1995, pioneering Ni phytomining trials were carried out at the U.S. Bureau of Mines on a naturally occurring stand of *Streptanthus polygaloides* A. Gray, a Ni-hyperaccumulating member of the Brassicaceae family endemic to serpentine soils in California (Reeves et al., 1981). The soil at the site contained a Ni concentration of 3340 mg kg⁻¹, in the normal range for serpentine soil. At the optimal harvest stage, the shoot Ni concentration of *S. polygaloides* averaged 5300 mg kg⁻¹, and biomass was 4.8 Mg ha⁻¹ with unfertilized plants (Nicks and Chambers, 1995). It was suggested that the potential value of a crop of Ni would be about the same as that of a crop of wheat, provided that an improved strain could be bred from selected plants and that some of the energy released by combustion of the dry material could be used to generate electricity. The economic analysis assumed that:

- the improved *S. polygaloides* strain contained 1% Ni in dry matter;
- biomass yield was 10 Mg ha⁻¹;
- price of nickel was \$7.65 kg⁻¹;
- a return to the grower of half of the \$765 value of the Ni;
- the energy of combustion of the biomass turned into electricity yielded \$131 ha⁻¹.

A later study by Robinson et al. (1997) in Italy using the well-known hyperaccumulator *Alyssum bertolonii* produced comparable economic data. In the last 3 years, the world price of nickel has ranged from \$5 to \$10 kg⁻¹. At the lower Ni prices, unless phytoextraction performance of naturally occurring hyperaccumulators is improved, Ni cannot be phytomined economically. Bennett et al. (1998) concluded that phytoremediation and phytomining techniques are still

in their infancy and will need to be proven over several years before there is complete scientific and commercial acceptance of their value.

In order to meet commercial phytoextraction requirements, our group has conducted large-scale field experimental research and laboratory studies over the past 6 years. A U.S. Patent has been granted on our phytomining proposal for specific metals including Ni (Chaney et al., 1998). Some of our results are discussed in this paper.

Economics of Ni phytoextraction

The first point we would like to make is that the hyperaccumulation trait is vital to the success of commercial phytomining of Ni. Phytoextraction efficiency depends both on high concentration of the target metals in harvestable shoot biomass and on biomass yield. Whether metal hyperaccumulation in shoots or high shoot biomass is more important in the phytoremediation of soil metals has been debated (Chaney et al., 1995; Cunningham et al., 1995; Salt et al., 1996). Chaney et al. (1997) gave a quantitative example to compare high biomass crops (such as *Z. mays* and *Brassica juncea*) with the Zn/Cd hyperaccumulator *Thlaspi caerulescens* J. Presl & C. Presl. In the case of the usual Zn and Cd co-contamination at 100 mg Zn:1 mg Cd, high biomass crops can only remove 5 kg of Zn ha⁻¹ year⁻¹. For *Thlaspi caerulescens* with a low yield of 5 tons ha⁻¹, Zn removal would be 125 kg ha⁻¹. Further, the biomass ash would contain 20–40% Zn in the case of *T. caerulescens*, but only 0.5% for *Z. mays* – the former is a rich ore, whereas the latter is a phytotoxic waste requiring disposal. Because a high metal concentration in the plant ash is important for efficient metal recovery, use of hyperaccumulators is crucial to the success of phytomining.

In recent field trials, we measured shoot Ni concentration as high as 22 g kg⁻¹ and biomass as high as 20 Mg ha⁻¹ in selected parental lines of *Alyssum murale* Waldst. & Kit. and *A. corsicum* Duby in our breeding program (unpublished data). Using data for the price of nickel over the past fifteen years (Figure 1), the economic potential of our technology for phytoextraction of Ni can be estimated as follows:

- 400 kg Ni ha⁻¹ (biomass yield of 20 Mg ha⁻¹ with biomass Ni concentration of 20 g kg⁻¹);
- \$7.33 kg⁻¹ Ni (1984–2001 average value);
- 25% of Ni value to cover cost of metal recovery and license and royalty fees;

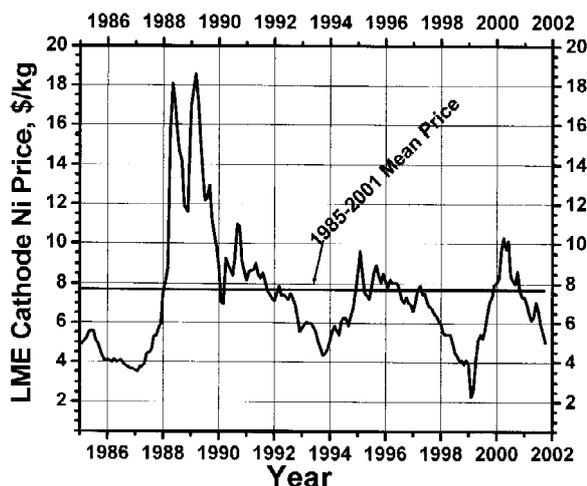


Figure 1. Variation in monthly mean cash price of cathode nickel on the London Metal Exchange (1985–2001).

- \$250 ha⁻¹ production costs;
- \$200 ha⁻¹ land rental cost;
- estimated net value of an annual phytomining crop would be \$1749 net ha⁻¹.

This estimation is rather conservative. Possible recovery of energy of combustion is not counted in the value, and it could offset some of the production costs. The nickel price is subject to cyclical variation. The biomass could be combusted immediately for its energy value and the plant ash stored until the world price improved. A bio-ore has much higher metal content than a conventional ore and therefore needs far less space for storage (Brooks et al., 1998). For phytoremediation of contaminated sites, there would be no land rental cost. U.S. Department of Agriculture data show that the average cash value of the U.S. wheat crop has been about \$300 ha⁻¹ in recent years. The annual value of grass hay, an alternative normal crop commonly grown on infertile serpentine soils, is about \$125 ha⁻¹.

Another potential source of income from phytomining is from the sale of carbon dioxide credits. Serpentine soils are naturally infertile, support little plant biomass, and, in some areas, have low levels of soil organic matter. Long-term cultivation of *Alyssum*, with use of fertilizers, should result in a permanent increase in soil organic matter levels and sequestration of atmospheric carbon dioxide.

Some observers have expressed concern that large-scale Ni phytomining will endanger the unique plant and animal species that are endemic to ultramafic and serpentine sites. We agree that it is important to pre-

Table 1. Comparison of Ni hyperaccumulator species grown in the greenhouse on Ni-contaminated agricultural soil and in the field (southwest Oregon) on serpentine soil

Experiment location	Soil	Species	Mean shoot Ni concentration (mg/kg)	Relative Size*
Greenhouse	Ni-contaminated agricultural	<i>Alyssum murale</i>	11300	L
		<i>Alyssum corsicum</i>	7270	L
		<i>Berkheya coddii</i>	5630	S
Field	Serpentine	<i>Alyssum corsicum</i>	18100	L
		<i>Alyssum murale</i>	15000	L
		<i>Alyssum pterocarpum</i>	13500	M+
		<i>Alyssum caricum</i>	12500	L
		<i>Alyssum bertolonii</i>	10900	M-

*S – small; M – medium; L – large.

serve biotic diversity, and we do not believe that the technology we are proposing is a threat to that diversity. A substantial proportion of serpentine areas are not farmable because of steep slopes, rocky soil, forest cover, or because they are within protected natural reserves. These areas will continue to be habitat for serpentine endemics. There are also large areas of serpentine soil that are currently being used for agriculture of low productivity – usually for grass hay or pasture – and it is these areas that are suitable for phytomining. The large acreage of arable serpentine soils and Ni-contaminated sites that exist worldwide will allow for potentially rapid growth of a Ni phytomining industry.

Developing a genetically improved hyperaccumulator crop

The commercial phytoextraction of Ni from polluted or Ni-rich soil requires development of a phytoextraction plant which combines the following characteristics:

- can be grown as an agricultural crop;
- grows rapidly and has high biomass;
- tolerates elevated concentrations of metals in soil;
- accumulates a significantly higher Ni concentration than exists in the soil.

To date, more than 300 Ni hyperaccumulating species have been described. Unfortunately, no plants that combine all the above properties have been identified in wild collections. The combination of the desirable characteristics of hyperaccumulator geno-

types through either classical plant breeding or genetic engineering is an important strategy for improving phytoextraction.

Understanding the genetic mechanism of metal accumulation in hyperaccumulator species is important for use of the molecular approach to genetic improvement. Schat et al. (2000) pointed out that transferring the hyperaccumulation syndrome into highly productive crop species by genetic engineering techniques seems to be far away. First, hyperaccumulation may be a complex phenomenon, because metal uptake, transport, and tolerance are apparently under independent genetic control. Second, none of the genes involved have been identified and cloned. In the absence of known ‘hyperaccumulation’ genes, attempts have been made to transfer the hyperaccumulation trait via somatic and sexual hybridization (Brewer et al., 1997).

Traditional plant breeding can use the available genetic diversity within a species to combine the traits needed for successful phytoextraction. Although there have been some efforts to study genotypic differences in metal uptake and biomass in hyperaccumulator species, little effort has been made to breed these species for domestication. Nicks and Chambers (1995) observed large variation in biomass within a *Streptanthus polygaloides* population in a field experiment. They found plants growing less than 20 cm apart to differ in size by a factor of two to three. Brooks and Robinson (1998) reported a very wide range of biomass, ranging from 1 to 64 g, in a *Thlaspi caerulescens* population growing over Zn/Pb mine wastes near Montpellier in southern France.

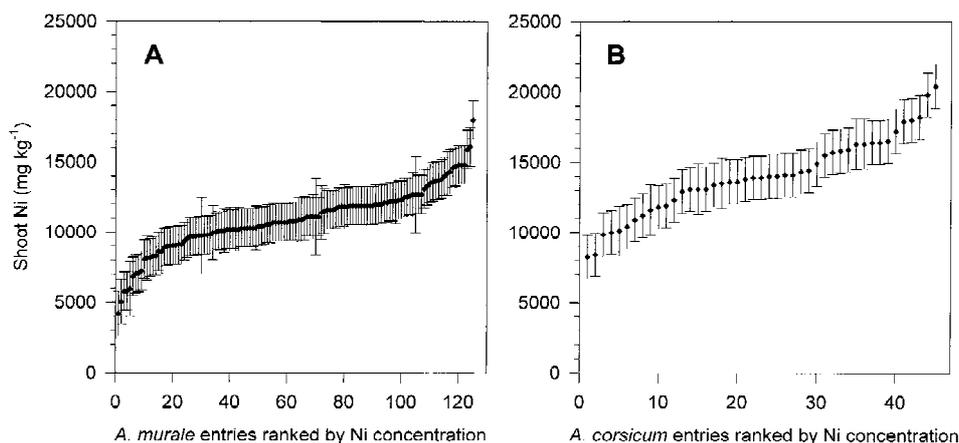


Figure 2. Mean shoot Ni concentration, with SE as bars, in (A) 125 *Alyssum murale* entries (B) 45 *Alyssum corsicum* entries grown on an Oregon serpentine soil.

In our studies on *Thlaspi caerulescens* growing on Zn smelter-contaminated soils at Palmerton, PA, 20 genotypes collected from Germany, Spain, France, UK, Belgium, Portugal and Norway were evaluated in a field experiment in 1996. After 1 year's growth, plant shoots were clipped for analysis of metal concentration. Significant differences were found among the 13 surviving genotypes for shoot Zn concentration. Field observations of plants clearly showed the wide range of biomass (Li et al., 2000).

In recent years, we have collected genetically diverse Ni hyperaccumulator species and ecotypes of *Alyssum*. Greenhouse pot tests and field evaluation were conducted using serpentine or Ni-refinery contaminated soils. In 1997, preliminary genetic screening experiments were conducted to identify Ni hyperaccumulator species and ecotypes suitable for further genetic improvement using both nutrient solution culture and field tests at a serpentine site in Baltimore, Maryland. Among 18 species and ecotypes tested, *Alyssum murale* Waldst. & Kit. and *A. corsicum* Duby had superior performance in comparison with others (Li et al., unpublished).

In 1998, we carried out a large genetic evaluation field experiment in Cave Junction, Oregon. The trial evaluated six selected Ni hyperaccumulator species, and included 125 *A. murale* and 45 *A. corsicum* accessions. The soil at the site is serpentine Brockman gravelly loam with a Ni concentration of 5500 mg kg⁻¹. DTPA-extractable Ni, a measure of the phytoavailable fraction, was 150 mg kg⁻¹. Large genetic variation was found in this study. The South African plant *Berkheya coddii* Roessl had the lowest biomass and shoot Ni concentration among the six Ni hyperac-

cumulator species tested (Table 1). This result was surprising because *B. coddii* has been suggested, over other Ni hyperaccumulators, as one of the better candidates for phytomining (Brooks and Robinson, 1998; Brooks et al., 1998). Our very different results may have been due to differences in climatic conditions. Within each hyperaccumulator species, large genetic variations for biomass and Ni shoot concentration were found in this study. Mean shoot Ni concentrations among *A. murale* and *A. corsicum* ranged from 4200 to 20400 mg kg⁻¹ (Figure 2). This finding indicates that genetic parental lines can be selected for breeding of improved phytoextraction cultivars. An on-going program is being carried out to combine superior characteristics and develop a genetically improved crop.

Ni phytoavailability studies were conducted on all soils used in these genetic evaluation experiments, as well as in all of the agronomic studies described below, using a variety of soil extractants, including diethylenetriaminepentaacetate (DTPA), to estimate Ni phytoavailability. It is widely known that DTPA-extractable soil Ni is positively correlated with plant Ni uptake. However, in our work with hyperaccumulators, we have found that the relationship is not a direct one, especially for field experiments. For example, some *Alyssum murale* accessions used in our large experiment at Cave Junction, Oregon, were also grown at another site in the Cave Junction area that had serpentine-influenced soil with a DTPA-extractable Ni concentration of only 25 mg kg⁻¹, one-sixth of the value for the soil in the main experiment. Shoot Ni concentration was decreased by an average of only 13% when *A. murale* was grown on the lower Ni

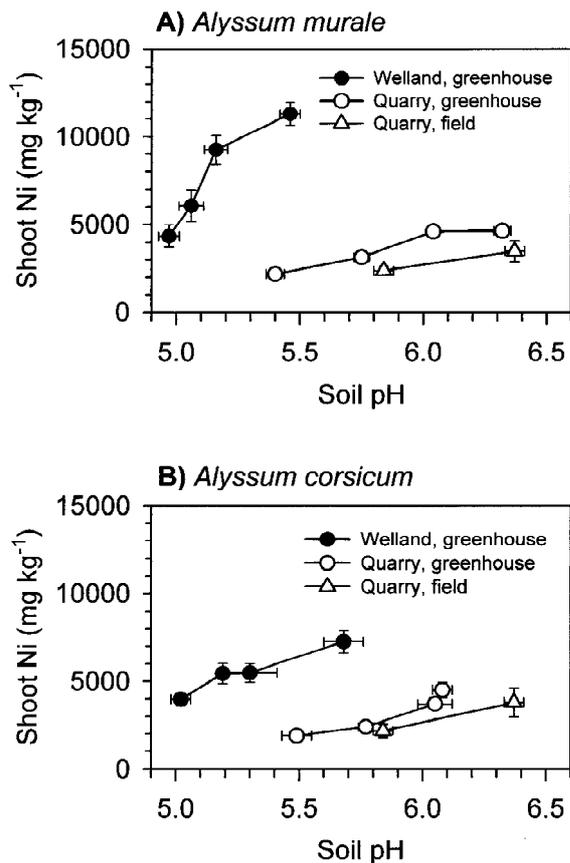


Figure 3. Effect of soil pH on shoot Ni concentration of two *Alyssum* species grown on two Ni-contaminated soils in greenhouse and field.

soil. Thus, the availability of soil Ni to at least some hyperaccumulators appears to be different from its availability to non-hyperaccumulator plant species.

Optimization of Ni uptake through soil management practices

Effective phytoextraction requires both plant genetic ability and the development of optimal agronomic management practices, including (1) soil management practices to improve the efficiency of phytoextraction; and (2) crop management practices to develop a commercial cropping system (Li et al., 2000).

Research has shown that modifying soil pH or soil fertility may affect the efficiency of phytoextraction of heavy metals such as Zn, Ni and Cd (Bennett et al., 1998; Brown et al., 1995a, b; Chiarucci et al., 1995; Robinson et al., 1997). However, different elements may be affected differently by pH, fertilization, and other management practices. Chaney et

al. (1998) pointed out that since soil pH is known to affect plant uptake of most heavy metals from soils, studies needed to be conducted to evaluate the independent effect of soil pH and soil metal concentration on hyperaccumulator yield and metal uptake.

We have been studying several soil management practices which may affect the efficiency of Ni phytoextraction. Soil pH is an important factor affecting absorption of metals by plants. In our studies, two Ni hyperaccumulator species, *Alyssum murale* Waldst. & Kit. and *A. corsicum* Duby, were used in both a greenhouse and a field experiment. For the greenhouse experiment, soils were collected at two sites near a historic Ni refinery in Ontario, Canada. The soils were a Quarry muck (Terric Mesisol) rich in organic matter and a Welland loam (Orthic Humic Gleysol). Both soils were contaminated by emissions; soil total Ni concentrations were 1700 mg kg⁻¹ for the Quarry muck soil and 2550 mg kg⁻¹ for the Welland loam. DTPA-extractable Ni concentrations were 480 mg kg⁻¹ for Quarry muck and 590 mg kg⁻¹ for Welland loam. Soil pH was adjusted using nitric acid and CaCO₃ to provide a range of pH before planting (Figure 3). Acidified treatments were leached to remove excess nitrate before fertilizers were added. A field study was conducted using similar soils and two pH levels: 'as is' and increased by limestone addition.

An unexpected result of both experiments was that Ni uptake by these *Alyssum* species was reduced at lower soil pH and increased at higher soil pH. This result is opposite to the response seen in crop plants and to the pattern of accumulation of other divalent metals (e.g., Mn and Zn) by these Ni-hyperaccumulator species. For *A. murale* grown on Welland loam for 120 days in the greenhouse, shoot Ni concentrations were 11 300 mg kg⁻¹ at pH 5.46, 9250 mg kg⁻¹ at pH 5.12, 6060 mg kg⁻¹ at pH 5.06 and 4340 mg kg⁻¹ at pH 4.97. On Quarry muck soil, shoot Ni concentrations of *A. murale* were 4630 mg kg⁻¹ at pH 6.32, 4600 mg kg⁻¹ at pH 6.04, 3140 mg kg⁻¹ at pH 5.75 and 2180 mg kg⁻¹ at pH 5.4. For *A. corsicum*, there was a similar effect of soil pH on Ni in shoots (Figure 3). Shoot Ni concentrations at 60 days were similar to the 120-day values (data not shown). DTPA-extractable Ni in the soils, measured at the end of the experiment, declined with increasing pH as expected. Thus we observed a seemingly paradoxical situation in which *Alyssum* plants accumulated higher concentrations of Ni in their shoots when the amount of phytoavailable Ni in the soil was lower. In the subsequent field study, we found that limestone treatment significantly increased Ni concentration in *Alyssum* shoots compared

to the control (Figure 3). In contrast to the results for Ni, shoot Mn and Zn showed increased uptake with lower soil pH in agreement with the literature for non-hyperaccumulator plants. Further research is needed to elucidate the mechanisms responsible for this unusual pattern of Ni accumulation by *Alyssum* shoots in response to soil pH.

In soil fertility management studies, we found that N application significantly increased shoot biomass yield, but did not affect shoot Ni concentration, and therefore total amount of phytoextracted Ni was increased (Chaney et al., unpublished). Recently, several other researchers have examined the question of whether the use of fertilizers to increase biomass is at the expense of lowered metal concentration. In a greenhouse experiment, Bennett et al. (1998) studied the effect of fertilization on three hyperaccumulator species, *Alyssum bertolonii*, *Streptanthus polygaloides* and *Thlaspi caerulescens*. They found that the addition of N fertilizer increases the biomass of all three species. There was no difference in biomass or metal uptake for varying concentrations of P. *Alyssum* and *Thlaspi* had a slight reduction in Ni and Zn concentration when biomass of each plant was increased using an N level of 100 mg kg⁻¹. The authors concluded that the trade-off of biomass against metal content was slight, but pointed out that the plants had a short growing period (20 weeks) in the pots, and that the concentrations of Ni and Zn obtained were appreciably lower than those found in wild plants. The experiments carried out by Robinson et al. (1997) were performed on naturally occurring populations of *Alyssum bertolonii* in Italy. Fertilization with nitrogen, phosphorus, and potassium more than doubled annual biomass production without reducing shoot Ni concentration. This is in agreement with our findings, which suggest that soil fertility management will be important for commercial phytoextraction.

Domestication of wild hyperaccumulator plant species into a useful crop

Domestication of a wild plant species into a commercially useful crop is usually a difficult and time-consuming undertaking. All aspects of cropping need to be evaluated in terms of the economics of phytoextraction using the improved crop. There have been few demonstrations of phytoextraction under field conditions (Li et al., 1997; McGrath et al., 1993; Nicks and Chambers, 1995). To date, few, if any, studies on phytoextraction have been devoted to develop-

ing crop management practices for improvement of phytoextraction efficiency.

We have carried out a number of field trials to study planting methods, population density, weed control practices, harvest schedule and methods, pollination control, and seed processing. Such crop management practice studies have improved phytoextraction efficiency and provide tools for farmers engaged in commercial production. Several important components of the crop management practices we have studied are described below.

Planting practices

The very small size and weight of the seeds of many hyperaccumulator species present a challenge to the agriculturalist. We first tested the effects of seeding depth, seedbed preparation, and light requirement for germination. The best results for some species, including *A. murale* and *A. corsicum*, were obtained by planting the seeds on the soil surface. Planting methods evaluated included (1) direct seeding, (2) seed pelletization, and (3) transplanting. Direct seeding is a straightforward and economical planting method. However, its drawbacks include the need for overseeding, the thinning of extra seedlings, and the risk of non-uniform stands. In addition, most hyperaccumulator species have slow seedling growth, and at that stage are very vulnerable to weed competition. Seed pelletization is widely used in horticulture to allow more convenient handling of small seeds. Unfortunately, pelletization resulted in poor germination of *Alyssum* seeds. Transplanting requires the cost and labor of growing seedlings in the greenhouse. Its advantages over other planting methods include: (1) uniform stand density, (2) plants get an earlier start in the growing season, and (3) reduced labor for weeding and thinning.

Planting density

Density (plants ha⁻¹) is expected to be an important management variable. The density of plants in a field usually influences the biomass yield per hectare and may affect metal concentration in the biomass. Little or no data has been published on this relationship for hyperaccumulator plants. From our field experiments, we found that the optimal planting density for commercial production is affected by soil physical properties, organic matter and fertility levels, hyperaccumulator species, and other factors (unpublished

data). On average, the optimal density for *A. murale* and *A. corsicum* is one plant per 0.25 m².

Annual/perennial management

A. murale and *A. corsicum* plants are perennial in nature. We are testing the effect of biomass harvest on the survival and subsequent growth of these species. It may be most economical to establish a perennial planting that is harvested every year. Alternatively, it may be more profitable to reseed each year or every other year. These choices could depend on the seasonal availability of rainfall or irrigation at production locations. Experimentation may have to be carried out in different regions of the world where Ni phytomining is conducted to evaluate whether annual, biennial, or perennial crop management is more cost-effective.

Weed control practices

Development of a new domesticated crop requires identification of weed control practices. Where labor cost is high as in the U.S. and Western Europe, hand weeding is not an option. Weed control chemicals are used to reduce weed competition with crops for soil nutrients, moisture, and light. We have conducted greenhouse and field tests of twelve herbicides for use with *A. murale* and *A. corsicum*, evaluating dose, mode of delivery, and use in combination. A strategy of weed control was developed that uses a combination of chemicals together with some mechanical weeding. Trifluralin (Treflan®) applied pre-plant and incorporated controls grasses and some broadleaf weeds. Postemergence, fluazifop (Fusilade®) can be used to control grasses. Pyridate (Tough®) can be used on established *Alyssum* plants (at least 8 weeks old) to control a number of broadleaf weeds. The following herbicides caused significant damage to *Alyssum*: clopyralid (Stinger®), oxyfluorfen (Goal®), pronamide (Kerb®), napropamide (Devrinol®), pendimethalin (Prowl®), dicamba (Banvel®), metsulfuron (Ally®), thifensulfuron+tribenuron (Harmony®), thifensulfuron (Pinnacle®), halosulfuron (Permit®), prosulfuron (Peak®), and bromoxynil (Buctril®). In serpentine soils, weed pressure is generally not as severe as it is in more fertile soils. More research is needed to find new chemicals in order to better control weeds in commercial production.

Harvest methods

The aboveground biomass of *Alyssum* can be harvested like 'hay' using standard farm machinery. The

plants are mowed when they are at the early flowering stage, generally in early June in the northern hemisphere. The biomass has a low moisture content and can usually be baled within two days of cutting. The bales can be stored in the field or near the biomass-to-energy facility.

Biomass processing and Ni recovery

Plant biomass must be processed to produce ash in a form that can be used as ore. We have done some work on developing efficient and cost-effective methods of Ni recovery. In a preliminary trial, the ash from our field biomass contained 32% Ni (unpublished data). Recovery of energy by biomass burning or pyrolysis could help make phytoextraction more cost-effective. Brooks and Robinson (1998) point out that a limitation on the use of an incinerator to produce steam for power generation is that hyperaccumulator crop harvesting would occur over a fairly short space of time, and therefore the power plant should be situated in an area where other waste might be used as a feedstock to keep the plant going the rest of the year.

Conclusion

Phytoextraction has several advantages over conventional remediation and mining processes. The use of genetically improved hyperaccumulator crops to decontaminate polluted soils is cost effective and could result in production of a bio-ore with commercial value that would help defray the costs of soil remediation. Carried out as we propose, phytomining would be an environmentally friendly method of obtaining metals important to industry, utilizing low-grade ores that otherwise would be unavailable for metal extraction.

Our R&D work as well as studies carried out by other researchers has contributed to a better understanding of the potential and limitations of Ni phytoextraction. The progress made in our recent studies will enable us to apply this technology commercially.

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