Is the bioavailability index applicable for trace elements in different types of soil?

Shuzhen Zhang and Xiao-quan Shan*

Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, PO Box 2871, Beijing 100085, China

* To whom all correspondence should be addressed: E-mail: xiaoquan@mail.rcees.ac.cn

ABSTRACT

The bioavailability index (BI) is defined as the proportion of reduction in a plant's accumulation of an element, caused by the removal of the extractable fraction of the element of interest from the soil. The BI and corresponding experimental methods were quantitatively applied to evaluate the bioavailability of trace elements in five Chinese soils. The soil was first extracted with various reagents (DTPA, HCl and $\text{NH}_2\text{OH}\cdot\text{HCl}$) separately, to remove the extractable elements. The soil, after extraction, was washed with deionised water to eliminate the extractant used in the fractionation analysis. Then the pH of the soil was re-adjusted with CaO. The soil was then fertilised and incubated in a greenhouse for four weeks. Tests showed that after incubation the pH, cation exchange capacity (CEC) and organic matter (OM) of the treated soil were close to their original values. Wheat (*Triticum aestivum* L.) was planted in both the untreated and treated soil for eight weeks. After harvest the plant concentrations of the elements Cr, Mn, Co, Ni, Zn, Cu, Cd, and Pb were analysed by inductively coupled plasma mass spectrometry (ICPMS). Trace element accumulation by plants grown in the treated soil was reduced significantly compared with that of plants grown in the untreated soil. The results showed that BI values were in the order $\text{BI(DTPA)} > \text{BI(HCl)} > \text{BI(NH}_2\text{OH}\cdot\text{HCl)}$. This indicated that the DTPA-extractable fraction represented a highly available fraction of the total content. Variations of BI among different trace elements show that Cr, Mn, Zn and Co have a higher BI, in general for the elements tested, whereas, Cu, Cd and Pb have lower values. There are also slight differences in the BI among soils. However, no significant relationship could be found between the BI and the soil characteristics.

Keywords: bioavailability index, speciation, soil, trace element.

INTRODUCTION

In recent years, many studies have been conducted to identify and evaluate the bioavailability of trace elements in soils, in order to assess the potential contamination of the food chain induced by soils (Maiz *et al.*, 1997; Quevauviller *et al.*, 1996; Kennedy *et al.*, 1997). In practice, most of the approaches estimate the amount of metal extracted from soils by various procedures, and correlate it with the metal content of plants. The correlation coefficients are normally used as the criterion of bioavailability. If plant accumulation of an element is significantly correlated with an extractable fraction, the fraction of the element is considered to be a good measure of plant bioavailability.

The extraction method and correlation analysis have some associated drawbacks, although they can still yield some useful information. Bioavailability depends on many factors such as soil properties and plant species. Unpredictable variations may greatly lower the degree of correlation and, in some cases, even cause a negative correlation of values between the extractable content of elements in soils, and the total content of the element in plants (Davies, 1992; Singh and Narwal, 1984; Alegria *et al.*, 1991). Alegria *et al.* (1991) found the highest correlation values between the total soil content and the uptake of Ni and Cd by vegetable species, whereas, the correlation between the extractable content of soils and the total content of Cd in leaves or stems was found to be negative. In other

research conducted by Singh and Narwal (1984) on Co, most of the extractants tested had a negative correlation with uptake by rape. For Ni, extraction by NH_4OAc (pH7.0) yielded negative correlation coefficients with Ni concentration in plant. It is difficult to interpret the negative correlation coefficients from the point of bioavailability. We cannot simply conclude that the extractants used are not adequate indicators of plant availability. On the other hand, the use of a fractionation method only provides empirical data, which are often not applicable to practical situations and natural field conditions, or only applicable to a narrow range of soil types. A final difficulty is related to the inherent nature of the statistical analysis itself, which necessitates a large number of soils, and it is better to use a wide range of metal concentrations to perform a reliable correlation analysis. Therefore, there is a need to develop a method, which could simply and reliably estimate plant availability as well as predict the uptake of trace metals by plants. Thus it would also evaluate the potential risk of these elements in contaminated environments.

Chen *et al.* (1996) presented a bioavailability index and the corresponding experimental method for the quantitative evaluation of plant availability of trace elements in soils. Since extractable trace elements generally represent the readily available fraction of the elements in soils, soils were first treated with an extractant, followed by pH adjustment and fertilisation. Crops were planted in both the treated and untreated soils. The BI was calculated as the following

$$\text{BI} = \frac{\Delta C_p / C_p^0}{C_e / C_t}$$

Where ΔC_p is the difference of the element content between the plants grown in the untreated and treated soils, C_p^0 is the element content in the plant grown in the untreated soil, C_e and C_t are the extractable content of the element and total concentration of the element in the soil, respectively. Therefore, the BI describes the proportion of the reduction of trace element uptake by plants which is attributable to the removal of the extractable fraction of the elements from soils. If removing a relatively small fraction of the total element content from the soil caused a significant reduction in the plant's accumulation of the element, a high BI value would be obtained. Consequently it could be concluded that this fraction was of high bioavailability.

The BI is a simple and a quantitative measurement for the bioavailability of different extractable fractions. However, in the previous study (Chen *et al.*, 1996) only two calcareous soils were tested. As we know, the physical and chemical characteristics of soils may influence

the bioavailability of trace metals in the soils. Therefore, further work with different soils is necessary. The aim of the present study is to investigate the application of BI to soils with different physico-chemical characteristics.

MATERIALS AND METHODS

Soils

Soil samples (0–20 cm) were collected from five different rural regions, stretching from northern to southern China. The soils, all of which were cultivated, varied widely in physico-chemical properties. The soil samples were air dried and ground to pass through a 2-mm sieve, homogenized and stored until analysis. Soil pH was measured in a 1:1 soil/water mixture. The organic matter was determined by the Walkley-Black method (Nelson and Sommers, 1982). The cation exchange capacity (CEC) was determined by the method described by Rhoades (1982). Total contents of trace elements were determined after samples were digested with $\text{HNO}_3/\text{HClO}_4/\text{HF}$ under high pressure (Zhang and Shan, 1997). The results are listed in Table 1.

Chen *et al.* (1996) described the soil treatment's experimental design. Briefly, soils were first extracted using selected extractants. The extraction procedures are given in Table 2. After extraction, the soils were washed with distilled-deionised water several times to eliminate the extractant. Then the soil pH was adjusted by adding a certain amount of CaO in order to restore the soil pH to its approximately original level. Triplicate samples were prepared at the same time.

For the pot experiments, 1 kg of both treated and untreated soils were placed in plastic pots. The pots received a supplemental fertilizer with the mixture solution of ammonium nitrate and potassium orthophosphate, supplying 10, 10, 13 mg/pot of elemental N, P and K, respectively. The soils were incubated in a greenhouse to keep a certain moisture content for 4 weeks prior to planting.

After soil incubation, winter wheat (*Triticum aestivum* L.) was sown at a rate of 20 seeds per pot, and subsequently thinned to 10 plants per pot after germination. Water losses were accounted for by daily additions of deionized water to maintain a consistent moisture content. The plants, both tops and roots, were harvested 8 weeks after germination. The samples were washed thoroughly with deionized water, placed in paper bags, and dried to a constant weight at 60°C for 24h. The dried tissues were finely ground for the analysis.

Instrumental analysis

The metal concentrations in soils and plants were determined by inductively coupled plasma mass spectrometry (ICP-MS) after digestion with $\text{HNO}_3/\text{HClO}_4/\text{HF}$ (Zhang and Shan, 1997). The extractant was analyzed

Table 1 Selected chemical and physical properties of soils used in the pot experiments

Property	Soil				
	Soil 1	Soil 2	Soil 3	Soil 4	Soil 5
Location	Guiyong	Jinhua	Yunnan	Heilongjiang	Beijing
Order	Entisol	Ultisol	Alfisol	Mollisol	Mollisol
Texture	Silty-clay, mixed	Fine-silty, mixed	Fine-silty, mixed	Sandy-silty, mixed	Sandy-silty, mixed
pH	7.52	6.19	7.10	6.28	7.50
CEC (me100g ⁻¹)	2.13	25.8	12.22	28.48	6.47
OM (%WW ⁻¹)	5.07	2.06	4.34	5.19	1.77
Total (mg kg ⁻¹)					
Cr	168.3	84.6	220.7	102.2	63.4
Mn	2870.9	1167.0	2851.7	1901.4	966.8
Co	33.1	18.5	29.4	16.4	10.2
Ni	49.5	22.2	63.9	33.4	18.1
Zn	184.1	140.9	418.6	96.9	80.6
Cu	51.5	22.1	261.9	26.2	20.2
Cd	0.76	0.14	0.31	0.61	0.33
Pb	25.3	25.6	81.4	25.2	23.6

Table 2 Extractant and procedure used for bioavailability index

Extractant	Composition	Soil/extractant ratio(w/v)	Shaking time
DTPA	0.005MDTPA+0.01M CaCl_2 +0.1M TEA(triethanolamine) pH7.3	1/10	2h
1MHCl	1.0 M HCl	1/10	2h
NH ₂ OH·HCl	0.05M NH ₂ OH·HCl+ 1M HCl	1/10	2h

after 5 dilutions. The ICP-MS was optimised with a solution of ^{115}In at 10 ng mL^{-1} in 2% HNO_3 , which was also used as an internal standard to compensate for matrix suppression and signal drifts during analysis. The accuracy of the elemental analysis was checked by the determination of REEs in the certified reference materials (GSS-1, GSS-8 of soils and GBW 07605 of tea from the National Research Center for Certified Reference Materials, Beijing, China) with seven replicate samples. The results are summarised in Table 3. Good agreement was achieved between the data obtained by the present method and the certified values for both soil and tea leaves.

RESULTS AND DISCUSSION

Effects of soil preparation upon soil properties

Previous investigations have indicated that bioaccumulation of trace elements is to some extent dominated by the soil properties (Soon and Bates, 1982; Sauerbeck and Hein, 1991; Davies, 1992; MaNeal *et al.*; 1985). If the soil properties have changed during the treatment procedures, the reductions of the trace elements in the plants would not only be controlled by the removal of trace elements, but also influenced by the changes in soil properties.

We investigated whether the pH of the treated soil can be adjusted to its original condition without affecting soil properties. The pH, CEC and OM of one soil sample were tested after the soil treatment. As shown in Table 4 there was no significant difference between the treated and untreated soils. Furthermore, care was taken to determine whether the soil treatment proce-

Table 3 Determination of trace elements in the certified reference materials by ICP-MS

Element	GSS soil ($\mu\text{g g}^{-1}$)		GBSS soil ($\mu\text{g g}^{-1}$)		GBW7605 tea leaves ($\mu\text{g g}^{-1}$)	
	This method*	Certified	This method*	Certified	This method*	Certified
Cr	58.5 ± 3.1	62 ± 6	70.4 ± 1.2	68 ± 8	1.5 ± 0.2	2
Mn	1801.7 ± 3.9	1760 ± 98	667.9 ± 4.1	650 ± 35	0.28 ± 0.03	0.217 ± 0.011
Co	15.0 ± 0.8	14.2 ± 1.5	11.4 ± 0.5	12.7 ± 1.7	0.21 ± 0.02	0.18
Ni	18.9 ± 1.0	20.4 ± 2.7	30.8 ± 1.1	31.5 ± 2.7	5.8 ± 0.4	5.09 ± 0.76
Zn	669 ± 4.1	680 ± 39	71.4 ± 1.6	68 ± 6	24.0 ± 1.8	22.6 ± 1.5
Cu	22.3 ± 0.8	21 ± 2	25.6 ± 2.0	24.3 ± 1.8	10.01 ± 0.2	8.96 ± 0.59
Cd	4.6 ± 0.2	4.3 ± 0.6	0.14 ± 0.02	0.13 ± 0.05	0.03 ± 0.001	0.023 ± 0.004
Pb	92.3 ± 1.9	98 ± 8	23.1 ± 1.2	21 ± 3	0.98 ± 0.01	$1.00 \pm 0.05^*$

Mean and standard deviation from 5 determinations.

Table 4 Selected properties of soil 5 before and after extraction treatment

	untreated	treated		
		DTPA	HCl	NH ₂ OH.HCl
pH	7.50	6.96	7.23	7.28
CEC me 100 ⁻¹	6.47	5.85	6.63	6.50
OM% W W ⁻¹	1.38	1.62	2.00	1.47

dures caused differences in plant growth. The experimental results indicated no significant differences between the yields of treated and untreated soils.

Comparison of BI among different extractants

Three different types of extractants, ammonium ethylenediaminetetra-acetate, hydrochloric acid and Hydroxyl-ammonium chloride were selected for soil extraction because they represent different kinds of chemical attack in addition to being commonly used to investigate the bioavailability of elements to plants (Maiz

et al., 1997; Quevauviller *et al.*, 1996; Lindsay and Norvell, 1978; Das *et al.*, 1995).

The BI values of the extractable trace elements with different extractants are listed in Tables 5(a) to 5(e) for different soils. For all the soils, the BI values were in the order of BI (DTPA>BI (HCl)>BI (NH₂OH.HCl). This suggested that DTPA-extractable trace elements represent a highly available fraction of the total content. When this highly available fraction of the elements was removed by DTPA extraction, the element accumulation by plants was significantly affected. This result corresponded to that of previous studies on DTPA as an extractant for assessing bioavailability of micronutrient cations in soils. Randall *et al.* (1976) concluded that DTPA could be used successfully to determine the available Mn in low organic matter soil. Bingham *et al.* (1975) showed a highly significant correlation between DTPA-extractable Cd and that taken up by corn. Singh and Narwal (1984) found numerically superior correlation coefficients between Cd, Pb and Zn extracted by DTPA and the total uptake of these metals by rape

Table 5 (a). BI of the extractable trace elements with different extractants for soil 1*

Element	BI of DTPA-extractable fraction					BI of HCl-extractable fraction					BI of NH ₂ OH.HCl extractable fraction				
	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R
Cr	0.80	25.1	31.4	21.9	27.4	3.2	25.6	8.0	32.0	10.0	7.2	26.8	3.7	49.6	6.9
Mn	1.0	34.4	34.4	31.6	31.6	14.6	69.2	4.7	46.7	3.2	28.4	41.7	1.6	56.8	2.0
Co	3.1	29.3	9.5	26.4	8.5	7.7	49.6	6.4	47.0	6.1	38.8	67.4	1.7	69.9	1.8
Ni	2.6	30.0	11.5	35.9	13.8	10.7	21.4	2.0	16.1	1.5	15.6	24.8	1.6	29.6	1.9
Zn	2.7	35.9	13.3	45.0	16.7	5.6	32.2	5.7	16.2	2.9	23.5	53.9	2.3	65.8	2.8
Cu	10.8	18.3	1.7	27.0	2.5	21.9	24.5	1.1	35.1	1.6	28.2	26.0	0.92	32.6	1.2
Cd	9.7	28.1	2.9	40.7	4.2	29.9	29.2	1.0	50.5	1.5	33.3	40.7	1.2	41.7	1.2
Pb	7.5	33.1	4.4	33.0	4.4	44.3	39.9	0.9	31.0	0.7	30.9	24.7	0.8	60.7	2.0

*The extractable trace elements (C_E) are expressed in percentage of the total content of the elements in the untreated soil and plant accumulation reduction of the elements in plant (ΔC_P) and in root (ΔC_R) are expressed in percentage of the total plant content of the elements in the untreated soil. Values of C_E, ΔC_P and ΔC_R listed in the table are means of the three replicates, BI for plant (BI_P) and for root (BI_R) are the ratio of ΔC_P to C_E and ΔC_R to C_E respectively.

Table 5 (b). BI of the extractable trace elements with different extractants for soil 2*

Element	BI of DTPA-extractable fraction					BI of HCl-extractable fraction					BI of NH ₂ OH.HCl extractable fraction				
	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R
Cr	0.52	25.6	49.2	57.0	109.6	3.0	39.9	13.3	52.6	17.5	8.2	23.0	2.8	64.6	7.9
Mn	0.58	20.2	34.8	34.9	60.2	7.6	35.7	4.7	37.5	4.9	19.0	55.9	2.9	60.8	3.2
Co	0.97	49.8	51.2	51.5	53.1	21.0	75.6	3.6	72.1	3.4	21.2	59.5	2.8	65.1	3.0
Ni	3.2	54.4	17.0	54.6	17.1	5.4	8.9	1.6	41.3	7.6	9.6	20.1	2.1	13.9	1.4
Zn	3.0	32.2	10.7	88.9	29.6	8.7	42.4	4.9	53.7	6.2	8.3	33.6	4.0	43.3	5.2
Cu	6.2	24.5	4.0	16.0	2.6	13.3	21.6	1.6	24.8	1.9	51	32	0.63	22.8	0.45
Cd	10.1	29.2	2.9	42.4	4.2	10.7	69.6	6.5	77.8	7.3	11.7	62.9	5.4	74.4	6.4
Pb	4.0	39.9	10.0	45.3	11.3	22.3	39.1	1.8	59.1	2.7	21.7	38.7	1.8	71.1	3.3

*The extractable trace elements (C_E) are expressed in percentage of the total content of the elements in the untreated soil and plant accumulation reduction of the elements in plant (ΔC_P) and in root (ΔC_R) are expressed in percentage of the total plant content of the elements in the untreated soil. Values of C_E, ΔC_P and ΔC_R listed in the table are means of the three replicates, BI for plant (BI_P) and for root (BI_R) are the ratio of ΔC_P to C_E and ΔC_R to C_E respectively.

Table 5 (c). BI of the extractable trace elements with different extractants for soil 3*

Element	BI of DTPA-extractable fraction					BI of HCl-extractable fraction					BI of NH ₂ OH.HCl extractable fraction				
	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)C _E	BI _R
Cr	0.75	43.2	57.6	24.7	33.2	2.2	33.9	15.4	25.5	11.6	7.1	41.2	5.8	61.2	8.6
Mn	0.80	30.0	37.5	38.0	47.5	17.7	51.1	2.9	1.2	44.2	25.9	44.0	1.7	62.3	2.4
Co	1.7	43.9	25.8	45.0	26.5	23.3	50.6	2.2	44.8	1.9	23.4	65.6	2.8	40.2	1.7
Ni	3.3	27.4	8.3	30.4	9.2	21.5	21.7	1.0	54.5	2.5	21.5	21.3	1.0	29.4	1.4
Zn	2.2	29.5	13.4	27.9	12.7	19.1	39.2	2.1	37.8	2.0	19.0	30.9	1.6	41.8	2.2
Cu	4.7	23.5	5.0	22.5	4.7	14.1	10.8	0.76	36.7	2.6	20.3	14.6	0.72	30.8	1.5
Cd	15.0	44.3	2.9	45.0	3.0	21.5	28.0	1.3	36.6	1.7	34.7	41.5	1.2	47.5	1.4
Pb	4.5	22.7	5.0	26.3	5.8	56.0	43.2	0.8	56.7	1.0	56.1	63.3	1.1	59.2	0.9

*The extractable trace elements (C_E) are expressed in percentage of the total content of the elements in the untreated soil and plant accumulation reduction of the elements in plant (ΔC_P) and in root (ΔC_R) are expressed in percentage of the total plant content of the elements in the untreated soil. Values of C_E, ΔC_P and ΔC_R listed in the table are means of the three replicates, BI for plant (BI_P) and for root (BI_R) are the ratio of ΔC_P to C_E and ΔC_R to C_E respectively.

Table 5 (d). BI of the extractable trace elements with different extractants for soil 4*

Element	BI of DTPA-extractable fraction					BI of HCl-extractable fraction					BI of NH ₂ OH.HCl extractable fraction				
	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)C _E	BI _R
Cr	0.37	25.7	69.4	28.0	75.7	1.3	12.6	9.7	16.9	13.0	6.3	29.0	4.6	21.6	3.4
Mn	1.27	57.3	45.1	76.3	60.0	3.6	34.1	9.5	59.8	16.6	27.9	56.2	2.0	53.2	1.9
Co	0.73	59.8	81.9	51.6	70.7	26.4	49.0	1.9	39.6	1.5	26.6	59.3	2.2	57.3	2.2
Ni	1.26	25.5	20.2	28.9	22.9	12.7	29.4	2.3	34.0	2.7	12.8	48.8	3.8	40.6	3.2
Zn	0.90	28.4	31.5	44.5	49.4	11.4	28.3	2.5	41.7	3.7	15.8	41.2	2.6	56.5	3.6
Cu	7.3	34.9	4.8	37.0	5.1	14.5	53.7	3.7	46.4	3.2	23.6	51.9	2.2	62.0	2.6
Cd	8.7	10.4	1.2	17.0	1.9	9.7	41.5	4.3	56.3	5.8	10.7	65.3	6.1	78.5	7.3
Pb	3.4	19.4	5.7	36.4	10.7	18.7	34.2	1.8	66.3	3.5	18.8	25.9	1.4	39.5	2.1

*The extractable trace elements (C_E) are expressed in percentage of the total content of the elements in the untreated soil and plant accumulation reduction of the elements in plant (ΔC_P) and in root (ΔC_R) are expressed in percentage of the total plant content of the elements in the untreated soil. Values of C_E, ΔC_P and ΔC_R listed in the table are means of the three replicates, BI for plant (BI_P) and for root (BI_R) are the ratio of ΔC_P to C_E and ΔC_R to C_E respectively.

Table 5 (e). BI of the extractable trace elements with different extractants for soil 5*

Element	BI of DTPA-extractable fraction					BI of HCl-extractable fraction					BI of NH ₂ OH.HCl extractable fraction				
	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)	BI _R	C _E (%)	ΔC _P (%)	BI _P	ΔC _R (%)C _E	BI _R
Cr	0.91	35.7	39.3	36.7	40.3	13.6	33.1	2.4	55.7	4.1	2.7	36.0	13.3	38.1	14.1
Mn	2.3	34.1	14.8	69.9	30.4	5.5	15.1	2.7	12.7	2.3	19.4	40.7	2.1	42.8	2.2
Co	7.7	49.6	6.4	83.3	10.8	8.5	35.9	4.2	43.2	5.1	25.9	47.7	1.8	43.2	1.7
Ni	1.6	16.7	10.4	22.7	14.2	11.1	17.2	1.5	21.1	1.9	11.1	22.4	2.0	20.0	1.8
Zn	2.0	70.1	35.0	82.4	41.2	16.6	26.8	1.6	45.1	2.7	16.6	73.3	4.4	78.0	4.7
Cu	5.7	23.5	4.1	21.1	3.7	7.4	13.7	1.9	14.1	1.9	27.5	44.0	1.6	33.0	1.2
Cd	12.4	22.9	1.8	26.0	2.1	11.5	39.1	3.4	50.5	4.4	31.8	54.0	1.7	60.1	1.9
Pb	3.2	13.1	4.1	15.7	4.9	18.3	35.5	1.9	34.5	1.9	18.3	48.9	2.7	45.8	2.5

*The extractable trace elements (C_E) are expressed in percentage of the total content of the elements in the untreated soil and plant accumulation reduction of the elements in plant (ΔC_P) and in root (ΔC_R) are expressed in percentage of the total plant content of the elements in the untreated soil. Values of C_E, ΔC_P and ΔC_R listed in the table are means of the three replicates, BI for plant (BI_P) and for root (BI_R) are the ratio of ΔC_P to C_E and ΔC_R to C_E respectively.

fodder. Also Maiz *et al.* (1997) achieved good results using DTPA as an extractant to determine the available metal fractions of Cd, Pb, Zn, Cu, Mn, Ni, Fe and Cr in soils. Although HCl and $\text{NH}_2\text{OH}\cdot\text{HCl}$ removed more of the metals, they had lower BI values. The HCl extractant dissolves some of the more insoluble minerals though an acid attack, has strong cation-exchange potential, and keeps the cations in solution by forming chloride complexes. Therefore, some unavailable fractions were extracted, leading to lower BI values. The $\text{NH}_2\text{OH}\cdot\text{HCl}$ extractant is commonly used in fractionation for the elements in reducible forms or excluded in the reducible Fe–Mn oxides (Shuman, 1985; Lake and Lester, 1984), which are thought to be less available for plants. The BI values for Cd in soils 2 and 4 are somewhat different from other soils. They are not in the order of $\text{BI (DTPA)} > \text{BI (HCl)} > \text{BI (NH}_2\text{OH}\cdot\text{HCl)}$. The average values of BI for DTPA-extractable Cd for soils 2 and 4 were 2.9 and 1.2 respectively, which were lower than those for HCl and $\text{NH}_2\text{OH}\cdot\text{HCl}$, which were 6.5 and 4.3 respectively for the HCl fraction, and 5.4 and 6.1 respectively for the $\text{NH}_2\text{OH}\cdot\text{HCl}$ fraction. This phenomenon was probably caused by the addition of CaO for the pH adjustment. It agrees with Andersson and Persson's observations (1984) that occasional increases were observed in the uptake of Cd after small amounts of CaO had been added. They suggested that the effect was caused by the addition of Ca, resulting in the removal of the exchangeable Cd from the soil colloids, thereby temporarily rendering it increasingly available to plants. Also we can see from Table 5(a) to 5(e) that the BI values for HCl and $\text{NH}_2\text{OH}\cdot\text{HCl}$ extractable Cd for soils 2 and 4 are higher than the BI values for other soils. The explanation could be from the differences of the physical properties of the soils. Soils 2 and 4 have higher cation exchange capacity than other soils (see in Table 1), and the cation exchange reactions in soils 2 and 4 are easier than in the other soils. Therefore only in soils 2 and 4, could we observe the increase of Cd uptake in plant after CaO was added.

Bioavailability of different trace elements

Variations of BI among different trace elements were also examined. An approximate estimate could be made from Table 5 that, in general, Cr, Mn, Zn and Ni had higher BI values. The mean values for the DTPA extractable fraction were 49.4, 33.3, 20.8 and 13.5, respectively. Whereas Cu, Cd and Pb possessed lower values of 3.9, 2.3 and 5.8, respectively. This corresponded to the results obtained by Chen *et al.* (1996), in which Mn, Ni, Cr and V had high BI values, Cu had a low BI value and Zn, Co and Pb in the middle range. It should be emphasised that the BI variations among different trace elements showed the same pattern for different soils. From this, we may ascertain that BI can be used for different soils as an index for the study of

bioavailability.

From the definition of BI we can see that two factors can determine the difference of BI values among the elements. One is the different extractability of elements. Data from Table 5 shows that Cr, Mn and Co had low C_e , but the corresponding BI values were high. On the contrary, Cu, Cd and Pb had high C_e values, but the BI was low. Different elements exist as different chemical forms in soils and their interactions with the extracts were not the same. For some elements, only the most available species, such as complex ions in soil solutions and easily exchangeable ions, were extracted. Although these species comprise a small proportion, they are the main source of the bioavailable fraction. Accordingly, high BI values were obtained. Another effect was the difference in accumulation and reduction in uptake of the elements in plants caused by the soil treatments. However, as can be found in Table 5, accumulation and reduction values ΔC_p did not drastically differ. This is understandable because plants can develop an effective mechanism of active uptake and transport to obtain metal ions from less available fractions in order to obtain nutrients for growing.

Relationship between the bioavailability index and soil properties

Data in Table 5 shows that the BI values did not differ significantly among the soils. But it was notable that for the DTPA extractable fraction, soils 2 and 4 had slightly higher BI values than for most of the elements, which could be attributed to the much higher CEC and lower pH than for the other soils. The higher CEC and lower pH of soils would stimulate the release of more trace elements during incubation and during the plant's growing processes, thus improving the uptake of the elements to the plant. However, it is unclear which factor is predominant between pH and CEC.

Although variations of BI values among the soils were observed, they are not clearly related to soil properties. Bioavailability index is a relative accumulation and reduction measure by comparing treated and untreated soil. That means the comparison is made within the same soil. Also we can see from Table 5 that the BI values for plants are not different from those for roots, which may also be attributed to relative BI values obtained between the treated and untreated samples of the same soil. Therefore it may be expected that quantitative differentiation among the extractants with respect to trace element bioavailability, or comparison of the bioavailability among elements, could be achieved while minimizing the influence of soil properties.

CONCLUSION

The results of this study indicated that BI and the

corresponding experimental method could be used to establish quantitative relationships between soil extractability and the uptake of trace elements by plants. Therefore, a quantitative measure could be established for the comparison and evaluation of bioavailability of different extractable fractions. As described above, the BI of the DTPA-extractable fraction was much higher than that of HCl and $\text{NH}_4\text{OH}\cdot\text{HCl}$ extractable fractions. This indicates that the DTPA extractable fraction represented a highly available fraction of the total content. Variation of BI among different trace elements mirrors the differences of bioavailability among the elements. In this study higher BI values for Cr, Mn, Zn and Co and lower ones for Cu, Cd and Pb were observed. This difference was mainly attributed to the differences in extractability among the elements.

The influence of soil properties upon the BI values was insignificant. Quantitative differentiation among the extractants with respect to trace element bioavailability or the comparison of bioavailability among elements, could be achieved based on the BI method for different types of soil while minimising the effects of soil properties.

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