Plant Soil (2014) 379:231–245
DOI 10.1007/s11104-014-2058-0

REGULAR ARTICLE

Soil chemical properties affect the concentration of elements (N, P, K, Ca, Mg, As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) and their distribution between organs of *Rumex obtusifolius*

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Received: 4 September 2013 / Accepted: 6 February 2014 / Published online: 26 February 2014

Abstract
Background and aims The ionome (elemental composition) of grassland species has rarely been studied at the level of individual organs and little is known about effects of soil chemical properties on the ionome. Using the model oxalate plant *Rumex obtusifolius,* we asked how its biomass production and the distribution of elements between its organs is affected by soil chemical properties.
Methods We established a pot experiment with *R. obtusifolius* planted in acidic non-contaminated control and in slightly acidic and alkaline soils anthropogenically contaminated by the risk elements As, Cd, Pb, and Zn. Both contaminated soils were untreated and treated by lime and superphosphate. We determined biomass production and the concentrations of elements in its organs.
Results Biomass production was negatively related to the mobility of micro- and risk elements. Restricted transport of micro- and risk elements from belowground organs into leaves was recorded in untreated contaminated soils. In both lime-treated soils and in superphosphate-treated alkaline soil, elevated transport of micro- and risk elements from belowground organs into leaves was recorded in comparison to untreated contaminated soils. The lowest concentrations of micro- and risk elements were recorded in stems and seeds, followed by belowground organs and leaves.
Conclusions *R. obtusifolius* is an As-, Cd-, Pb-, and Zn-excluder and is sensitive to high availability of micro- and risk elements in the soil. Soil chemical properties affect the distribution of essential elements within the plant greatly.

Keywords Bioaccumulation and translocation factors · Broad-leaved dock · Oxalate plants · Quick lime · Superphosphate

Responsible Editor: Philip John White.

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Introduction

The ionome is the elemental composition of cells, tissues, organs or whole organisms (Salt et al. 2008). The elemental composition of many agricultural crops has been investigated (Šrek et al. 2012; Zhao et al. 2013). In the case of grassland species, the ionome has predominantly been studied in aboveground organs (White et al. 2012; Lindström et al. 2013). Therefore, insufficient information is available concerning the elemental composition of belowground organs. Even in aboveground organs, the elemental composition of grassland species is frequently determined only in bulk biomass or in leaves of individual species, to determine their forage quality or nutritional status (Thompson et al. 1997;

Hejcman et al. 2012a). Hence, little information is available to describe the distribution of elements in individual aboveground organs, particularly in the case of micro- and risk elements (Barman et al. 2000; Guleryuez et al. 2008; Gaweda 2009).

It has long been recognised that the elemental composition of plant tissues is influenced by environmental conditions and in particular by the availability of different elements in the soil (Barman et al. 2000; Anton and Mathe-Gaspar 2005). However, it is still not well known to what degree the concentrations of elements vary in different organs and how much of this variability is determined by soil chemical properties. Using the model oxalate plant *Rumex obtusifolius* subsp. *obtusifolius* (broad-leaved dock), which is a common weedy species in temperate grasslands (Hann et al. 2012; Strnad et al. 2012; Hujerová et al. 2013), the aim of this study was to test the extent to which concentrations of macroelements (N, P, K, Ca, and Mg), microelements (Cu, Fe, Mn, Ni, and Zn) and risk elements (As, Cd, Cr, Pb, and Zn) in its organs are affected by soil chemical properties. Zinc can be classified as both a microelement and a risk element, depending upon its availability in the soil and its concentration in plant biomass. *R. obtusifolius* belongs to the group of 'oxalate plants', which regulates excessive Ca concentrations in tissues by Ca-oxalate precipitation (White and Broadley 2003). Organic acids play an important role in heavy metal(-loid) tolerance and detoxification in plants because of their external or internal chelation with risk metal(loid)s (Sytar et al. 2013). In oxalate plants with low exudation rates of di- and tri-carboxylic acids (Tyler and Ström 1995), the main detoxification mechanism is probably internal chelation. The uptake and transport of micro- and risk elements by plants can be characterised by the bioaccumulation factor (BF), which is calculated as the plant-to-soil concentration ratio of a particular micro- or risk elements (Zhuang et al. 2007; Gupta et al. 2008). The leaf-to-root concentration ratio of particular micro- or risk elements is termed the translocation factor (TF; Gupta et al. 2008; Barrutia et al. 2009). According to Baker (1981), plants that accumulate micro- and risk elements are characterised by a BF and TF > 1, indicator plants by a BF and TF = 1 and plants that exclude these elements by a BF and TF < 1. Questions that have not yet been addressed are which BF and TF values are characteristic for oxalate plants like *R. obtusifolius* and to what extent are BF and TF values affected by the micro- and risk elements availability in the soil.

In this study, we asked how (1) biomass production of *R. obtusifolius*, (2) concentrations of elements in its organs and (3) BF and TF were affected by soil chemical properties.

Materials and methods

Design of the experiment

Two long-term heavily contaminated soils ('Litavka' by As, Cd, Pb, and Zn and 'Malín' by As, Cd, and Zn) and one control soil without any contamination ('Mšec') were used for a pot experiment in an outdoor university vegetation hall in Prague–Suchdol, with natural temperature and light conditions. Details concerning the history and sources of Litavka and Malín soil contamination are given in Borůvka et al. (1996) and Horák and Hejcman (2013). The physicochemical properties of all used soils are provided in Table 1.

In contaminated soils, we manipulated the availability of elements by application of lime and superphosphate according to previous findings of Vondráčková et al. (2013, 2014). We applied 7.3 g lime (CaO) per 1 kg of soil containing 686 g Ca kg^{-1} with pH_{CaCl2} 12.0 ±0.01 and 1.3 g superphosphate ($Ca(H_2PO_4)_2 \cdot H_2O$) per 1 kg of soil containing 246 g P kg^{-1} and 159 g Ca kg^{-1} with pH_{CaCl2} 2.2±0.003. The pot experiment was established in May 2011 with seven treatments replicated five times: LC—Litavka control soil without any additive; LCa—Litavka soil with lime; LP—Litavka soil with superphosphate; MC—Malín control soil without any additive; MCa—Malín soil with lime; MP—Malín soil with superphosphate; and McC—Mšec, non-contaminated control soil. Five kg of air dried soil were passed through a 10 mm sieve and put in 5-L pots (20 cm in diameter and height). In each pot, the whole soil profile was mixed with nutrient solution, consisting of 0.5 g N as NH_4NO_3, 0.16 g P and 0.4 g K as K_2HPO_4. Application of nutrient solution was performed, to ensure that N, P, and K availability was non-limiting for growth of *R. obtusifolius* in all treatments. The lime and superphosphate additives were mixed with the soil after application of the nutrient solution. One hundred seeds of *R. obtusifolius* were sown (1–2 cm

Table 1 Basic characteristic of the experimental soils (mean ± SE; n=3). Differences between soils, calculated by Kruskal–Wallis ranks, soils with the same letter were not significantly different test, were not statistically significant ($^{n.s.}$) or significant at 0.05 (*) probability level. Using the multiple comparisons of mean

Soil property	Soil		
	Litavka (49°43′N, 14°0′E)	Malín (49°58′N, 15°17′E)	Mšec (50°12′N, 13°51′E)
Soil texture	Clay loamy sand	Loam	Loam
Soil type	Fluvisol	Luvisol	Pararendzina
pH_{CaCl2}*	6.5 ± 0.02^{ab}	7.3 ± 0.02^{a}	5.2 ± 0.2^{b}
CEC ($mmol_{(+)}$ kg^{-1})*	109 ± 38^{ab}	333 ± 15^{a}	64 ± 12^{b}
C_{org} (%)*	3.6 ± 0.1^{a}	2.7 ± 0.1^{ab}	2.4 ± 0.2^{b}
P [a] (mg kg^{-1})*	9 ± 0.3^{a}	56 ± 3^{a}	175.5 ± 26.5^{a}
K [a] (mg kg^{-1})*	192 ± 8^{a}	234 ± 4^{a}	155 ± 3^{a}
Ca [a] (mg kg^{-1})$^{n.s.}$	1856 ± 31^{a}	8914 ± 98^{a}	1736 ± 216^{a}
Mg [a] (mg kg^{-1})$^{n.s.}$	160 ± 5^{a}	354 ± 5^{a}	117.5 ± 17^{a}
As $_{total}$ (mg kg^{-1})*	354 ± 2^{ab}	688 ± 26^{a}	9 ± 0.3^{b}
Cd $_{total}$ (mg kg^{-1})*	53.8 ± 0.9^{a}	11.3 ± 0.2^{ab}	0.2 ± 0.03^{b}
Cr $_{total}$ (mg kg^{-1})*	51.5 ± 0.8^{a}	45 ± 1^{ab}	18 ± 1^{b}
Cu $_{total}$ (mg kg^{-1})*	61 ± 0.4^{a}	62 ± 2^{a}	13 ± 1^{a}
Fe $_{total}$ (mg kg^{-1})*	21193 ± 146^{a}	17379 ± 224^{ab}	8469 ± 247^{b}
Mn $_{total}$ (mg kg^{-1})*	2688 ± 16^{a}	371 ± 4^{ab}	349 ± 17^{b}
Ni $_{total}$ (mg kg^{-1})*	18.5 ± 0.1^{ab}	23.5 ± 0.3^{a}	7 ± 1^{b}
Pb $_{total}$ (mg kg^{-1})*	3305 ± 85^{a}	98 ± 31^{ab}	32 ± 2^{b}
Zn $_{total}$ (mg kg^{-1})*	6172 ± 42^{a}	1022 ± 18^{ab}	40 ± 5^{b}

$_{total}$—pseudo-total concentrations of elements extracted by *Aqua Regia*

Czech legislation limits for pseudo-total concentrations of elements in light-textured/other soils (mg kg^{-1}): As 30/30, Cd 0.4/1.0, Cr 100/200, Cu 60/100, Fe not specified (n.s.), Mn n.s., Ni 60/80, Pb 100/140, Zn 130/200 (Anonymous 1994)

CEC cation exchange capacity (Schwertfeger and Hendershot 2009)

[a] Available concentrations of macro elements determined by Mehlich III extraction procedure (Mehlich 1984)

deep) in each pot (see Hejcman et al. 2012b for details about emergence and survival of seedlings in contaminated soils) and after 1 month of growth, the seedlings were thinned to three plants per pot. The pots were regularly watered with deionised water to maintain the optimal moisture condition for plant growth during the vegetative period. Positions of pots were changed weekly to avoid any side effect on the collected data. The plants were harvested after a growth period of 6 months and their biomass divided into belowground organs, stems, leaves, and seeds (i.e., achenes with a perianth). The belowground organs were first washed thoroughly with tap water to remove soil adhered to belowground organs. Then, the belowground organs were washed in ultrasound-assisted bath filled with deionised water (ELMASONIC S30, Elma Ultrasonic Technology).

Chemical analyses

Soil samples were collected from the whole soil profile at the end of the experiment and were analysed for pH, plant-available and acid-extractable concentrations of elements (Table 2). For chemical analyses, soil samples were air dried at 25 °C and sieved to <2 mm. Soil pH was measured in a 1:5 (w/v) suspension of soil and 0.01 mol L^{-1} $CaCl_2$. Mobile (plant-available) and mobilisable (acid-extractable) portions of elements in soils were determined using 0.01 mol L^{-1} $CaCl_2$ and 0.11 mol L^{-1} CH_3COOH (hereafter abbreviated as Ca and AA, Tlustoš et al. 1994; Quevauviller 1998). The

Table 2 Effect of treatment on soil pH, plant-available (mg kg^{-1}; extracted by 0.01 mol L^{-1} $CaCl_2$; Ca) and acid-extractable (mg kg^{-1}; extracted by 0.11 mol L^{-1} CH_3COOH; AA) concentrations of elements (mean ± SE) at the end of the experiment

Variable	Extraction	Treatment						
		LC	LCa	LP	MC	MCa	MP	McC
pH_{CaCl2}**	–	5.8±0.01^{c}	7.5±0.01^{ab}	5.9±0.02^{bc}	7.2±0.03^{ac}	7.6±0.02^{a}	7.2±0.01^{ac}	5.2±0.2^{c}
P **	Ca	1.0±0.2^{b}	1.1±0.2^{b}	3.4±0.6^{a}	1.5±0.1^{ab}	1.9±0.2^{ab}	3.3±0.1^{a}	4.2±0.6^{a}
K **	Ca	246±7^{a}	44±8^{b}	205±17^{a}	122±4^{ab}	90±2^{ab}	111±3^{ab}	44±9^{b}
Mg **	Ca	54±2^{ab}	23.5±1^{b}	50±3^{ab}	67±2.5^{a}	42±1^{b}	67±2.5^{a}	80±9^{a}
As **	Ca	0.35±0.04^{b}	0.33±0.1^{b}	0.4±0.1^{ab}	1.1±0.1^{ab}	1.4±0.1^{a}	1.8±0.1^{a}	0.05±0.02^{b}
Cd **	Ca	4.2±0.2^{a}	0.1±0.01^{ab}	3.8±0.2^{a}	0.02±0.002^{b}	0.03±0.01^{b}	0.04±0.01^{b}	0.03±0.004^{ab}
Cr $^{n.s.}$	Ca	0.03±0.004^{a}	0.03±0.01^{a}	0.03±0.01^{a}	0.02±0.01^{a}	0.03±0.01^{a}	0.02±0.003^{a}	0.01±0.005^{a}
Cu **	Ca	0.1±0.01^{ab}	0.3±0.02^{a}	0.1±0.01^{ab}	0.1±0.004^{b}	0.2±0.005^{a}	0.1±0.01^{b}	0.05±0.005^{b}
Fe **	Ca	7.5±0.9^{ab}	6.3±1.6^{b}	7.8±1.2^{ab}	11.2±1.4^{ab}	18.1±1.4^{a}	13.3±1.2^{a}	4.3±0.6^{b}
Mn **	Ca	9.1±0.5^{a}	0.3±0.03^{abc}	7.8±0.5^{abc}	0.2±0.02^{c}	0.2±0.02^{bc}	0.2±0.03^{abc}	10.4±1.9^{ab}
Ni **	Ca	0.2±0.003^{a}	0.04±0.01^{ab}	0.1±0.01^{a}	0.03±0.01^{b}	0.04±0.01^{ab}	0.05±0.01^{ab}	0.1±0.04^{ab}
Pb **	Ca	0.65±0.1^{a}	0.5±0.1^{a}	0.65±0.1^{a}	0.04±0.01^{b}	0.02±0.002^{b}	0.06±0.01^{ab}	0.1±0.02^{ab}
Zn **	Ca	179±7^{a}	3.7±0.7^{abc}	174±7^{ab}	0.6±0.1^{c}	1.3±0.3^{bc}	1.4±0.6^{c}	3.2±0.5^{abc}
P **	AA	3.1±0.5^{b}	3.2±0.4^{b}	18±4^{ab}	14±3^{ab}	11±0.5^{ab}	38±2^{a}	49±6^{a}
K *	AA	314±21^{a}	463±234^{ab}	346±56^{ab}	244±3^{ab}	237±5^{ab}	237±5^{ab}	75±13^{b}
Ca **	AA	1789±144^{b}	6484±1122^{ab}	2170±278^{b}	7991±155^{ab}	10779±191^{a}	8028±195^{ab}	1000±110^{b}
Mg **	AA	108±5^{ab}	129±18^{ab}	109±8^{ab}	559±15^{a}	515±14^{ab}	549±11^{a}	91.5±11^{b}
As **	AA	1.0±0.2^{bc}	1.1±0.05^{bc}	1.4±0.3^{abc}	7.9±0.7^{ab}	7.6±0.2^{ab}	14.5±0.7^{a}	0.6±0.1^{c}
Cd **	AA	27±1^{a}	26±2^{a}	27±1^{a}	3.0±0.1^{ab}	3.2±0.1^{ab}	2.9±0.1^{ab}	0.1±0.01^{b}
Cr **	AA	0.2±0.01^{abc}	0.3±0.03^{a}	0.25±0.02^{ab}	0.1±0.02^{bc}	0.1±0.02^{abc}	0.1±0.04^{abc}	0.04±0.002^{c}
Cu **	AA	1.8±0.1^{a}	2.1±0.4^{a}	1.8±0.1^{a}	0.7±0.03^{ab}	0.8±0.01^{ab}	0.7±0.03^{ab}	0.2±0.01^{b}
Fe **	AA	50±7^{ab}	41±6^{ab}	41±6^{ab}	59±7^{ab}	74±5^{a}	79±16.5^{a}	8.6±0.5^{b}
Mn **	AA	160±8^{a}	163.5±15^{a}	142±8^{a}	67±2^{b}	75±3^{ab}	64.5±1^{b}	76±1^{ab}
Ni **	AA	2.4±0.1^{a}	2.2±0.2^{a}	2.5±0.1^{a}	1.5±0.1^{ab}	1.3±0.1^{b}	1.5±0.03^{ab}	0.7±0.1^{b}
Pb **	AA	68±4^{a}	67±8^{ab}	52±3^{ab}	0.1±0.03^{c}	0.9±0.1^{abc}	0.3±0.1^{abc}	0.3±0.03^{bc}
Zn **	AA	2593±68^{a}	2284±114^{ab}	2607±76^{a}	270±5^{abc}	230±10^{bc}	272±5^{abc}	14±2^{c}

Treatment abbreviations: *LC* Litavka control soil without any additive, *LCa* Litavka soil with lime, *LP* Litavka soil with superphosphate, *MC* Malín control soil without any additive, *MCa* Malín soil with lime, *MP* Malín soil with superphosphate, and *McC* Mšec, non-contaminated control soil. Calculated by Kruskal-Wallis test, differences between treatments were not statistically significant ($^{n.s.}$) or were significant at 0.05 (*) and 0.01 (**) probability levels. According to the multiple comparisons of mean ranks, treatments with the same letter were not significantly different

element concentrations in soil extracts were determined using inductively coupled plasma-optical emission spectrometry (ICP-OES, VARIAN Vista Pro, Varian, Australia).

Fresh biomass was air-dried at 60 °C to total desiccation, dry matter biomass was determined and then plant samples were ground using a stainless-steel mill and subsequently analysed. The total concentrations of elements in organs were determined by ICP-OES (for P, As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) and flame atomic absorption spectroscopy (FAAS, VARIAN SpectrAA-280, Australia, for K, Ca, and Mg) after microwave-assisted high-pressure acid-digestion (65 % HNO_3:30 % H_2O_2 4:1, Ethos 1, MLS GmbH, Germany). Certified reference material (CTA-OTL-1 oriental tobacco leaves) was mineralised under the same conditions for quality assurance of the total element concentrations in experimental plants. The concentration of N in the plant organs was determined by the Kjeldahl method using a Vapodest 50s (Gerhardt, Königswinter, Germany) after wet-digestion with concentrated H_2SO_4 (98 %).

Data analysis

All statistical analyses were performed using the Statistica 9.0 (www.statsoft.com) and CANOCO 4.5 (ter Braak and Smilauer 2002) programs. All data were checked for homogeneity of variance and normality (Levene and Shapiro-Wilk tests). Soil and biomass data did not meet assumptions for the use of ANOVA and were thus evaluated by non-parametric Kruskal-Wallis test. We assessed the effects of 1) treatment on the concentrations of elements in the soil and biomass and on the BFs and TFs, 2) organ on concentration of elements in the biomass and 3) soil on the soil properties. After obtaining significant results from the Kruskal-Wallis test, we used multiple comparisons of mean ranks for the detection of significant differences between different soils, treatments or organs. The relationship between concentrations of elements in the biomass and biomass production was analysed by linear regression. A principal component analysis (PCA), in the CANOCO 4.5 program, was applied to all collected data together (concentrations of elements in the soil and biomass, pH, and biomass of organs). We used standardised 'species data' because data of different character and units were analysed together. The PCA was used to make visible correlations between all analysed data and similarity of different treatments. The results were visualised in the form of a bi-plot ordination diagram in the CanoDraw program.

Results

Biomass production

The effect of treatment on the total biomass of *R. obtusifolius* was significant (Fig. 1), and the total biomass weight ranged from 1.3 to 43.3 g plant^{-1} in the LP and the McC treatments, respectively. Belowground biomass was also significantly affected by the treatment, and ranged from 0.4 to 29.0 g plant^{-1} in the LC and the McC treatments, respectively (Table 3). Biomass production of all aboveground organs together was also significantly influenced by treatment. No stems or seeds were produced in the LC and LP treatments. Leaf biomass ranged from 0.1 to 5.8 g plant^{-1} in the LC and the McC treatments. In treatments with stems and seed production, stem biomass ranged from 1.1 to 4.1 g plant^{-1} in the MC and the LCa treatments, and seed biomass ranged from 1.9 to 7.9 g plant^{-1} in the MC and the LCa treatments, respectively. Significant negative relationships were recorded between concentrations of micro- and risk elements and total biomass of all organs in the case of Cd (r=−0.606; p<0.01), Ni (r=−0.373; p=0.033), Pb (r=−0.356; p=0.042), and of Zn (r=−0.552; p<0.01).

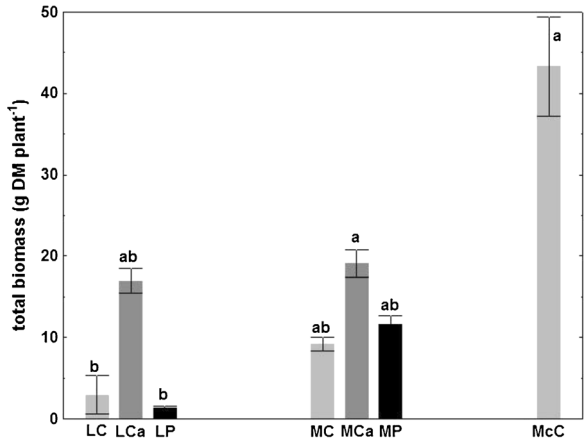

Fig. 1 Effect of treatment on the total biomass of *R. obtusifolius* (below- and aboveground biomass together) at the end of the experiment. Treatment abbreviations: *LC* Litavka control soil without any additive, *LCa* Litavka soil with lime, *LP* Litavka soil with superphosphate, *MC* Malín control soil without any additive, *MCa* Malín soil with lime, *MP* Malín soil with superphosphate, and *McC* Mšec, non-contaminated control soil. *Error bars* represent SE. Based on Kruskal–Wallis test, differences between treatments were significant based on a 0.01 (**) probability level. Using the multiple comparisons of mean ranks, treatments with the same letter were not significantly different

Concentration of macroelements in the organs

The concentrations of N, P, and Ca were significantly affected by treatments, and the concentrations of N, P, K, Ca, and Mg differed between individual organs (see Table 3 for details). The concentration of N ranged from 3.8 g kg^{-1} in stems in the MCa treatment, to 29.9 g kg^{-1} in leaves in the LP treatment. The concentration of P ranged from 0.3 g kg^{-1} in stems in the LCa treatment to 3.5 g kg^{-1} in leaves in the MC treatment; the concentration of K ranged from 5.6 g kg^{-1} in belowground organs to 38.9 g kg^{-1} in leaves in the LC and McC treatments, respectively, and the concentration of Ca ranged from 2.8 g kg^{-1} in stems to 15.4 g kg^{-1} in leaves both in the McC treatment. The concentration of Mg lay between 0.8 g kg^{-1} in stems in the McC treatment and 5.7 g kg^{-1} in leaves in the McC treatment.

Table 3 Effect of treatment on organ biomass (belowground organs, stems, leaves, and seeds) and total concentrations of elements (mean ± SE) in organs of *R. obtusifolius* in Litavka, Malín and Mšec soils. Treatment abbreviations: *LC* Litavka control soil without any additive, *LCa* Litavka soil with lime, *LP* Litavka soil with superphosphate, *MC* Malín control soil without any additive, *MCa* Malín soil with lime, *MP* Malín soil with superphosphate, and *McC* Mšec, non-contaminated control soil. Differences between treatments and organs were evaluated by Kruskal–Wallis test. For each element, concentrations in organs within one treatment denoted with the same letter (a–c) and concentrations in treatments within one organ denoted with the same letter (A–D) were not significantly different

Variable	Treatment	Organ			
		Belowground organs	Stems	Leaves	Seeds
Organ biomass (g plant^{-1})	LC	0.4 ± 0.2^{aC}	–	0.07 ± 0.03^{aC}	–
	LCa	11.5 ± 1.4^{aA}	4.1 ± 1^{abA}	3.9 ± 0.6^{bA}	7.9 ± 1^{abA}
	LP	1 ± 0.3^{aBC}	–	0.4 ± 0.2^{aBC}	–
	MC	6.6 ± 1.2^{aAB}	1.1 ± 0.2^{bA}	1.5 ± 0.3^{bABC}	1.9 ± 0.5^{abA}
	MCa	14 ± 2.5^{aA}	2.5 ± 0.8^{bA}	3.4 ± 0.6^{bA}	4.9 ± 0.9^{abA}
	MP	9.1 ± 1.3^{aA}	1.3 ± 0.6^{bA}	1.7 ± 0.3^{bAB}	2.4 ± 0.3^{abA}
	McC	29 ± 9^{aA}	3.6 ± 0.4^{aA}	5.8 ± 2.2^{aA}	5.1 ± 1.4^{aA}
N (g kg^{-1})	LC	23.3 ± 0.5^{A}	–	–	–
	LCa	10.3 ± 1.5^{abAB}	4.1 ± 0.2^{bA}	17.8 ± 1.4^{aB}	17.1 ± 0.6^{aA}
deficient	LP	22.7 ± 1.3^{bA}	–	29.9 ± 1.5^{aA}	–
normal 20–50[1]	MC	7.8 ± 0.9^{abAB}	3.95 ± 0.5^{bA}	18.1 ± 1.3^{aAB}	20.0 ± 0.6^{aA}
phytotoxic	MCa	8.3 ± 1.8^{abAB}	3.8 ± 0.3^{bA}	15.8 ± 1.7^{aB}	17.99 ± 0.95^{aA}
	MP	6.6 ± 0.3^{aAB}	5.4 ± 0.2^{aA}	18.1 ± 1.3^{aB}	17.6 ± 1.1^{aA}
	McC	5.7 ± 0.3^{aB}	6.1 ± 0.9^{aA}	22.8 ± 0.8^{aAB}	18.4 ± 1.0^{aA}
P (g kg^{-1})	LC	0.6 ± 0.04^{bB}	–	2.3 ± 0.8^{aAB}	–
	LCa	0.96 ± 0.05^{abAB}	0.3 ± 0.03^{bB}	2.1 ± 0.2^{aAB}	1.5 ± 0.3^{aB}
deficient <2(1)[1]	LP	0.97 ± 0.09^{aAB}	–	1.3 ± 0.2^{aB}	–
normal 3–5[1]	MC	2.0 ± 0.3^{abA}	0.8 ± 0.15^{bAB}	3.5 ± 0.9^{aAB}	2.8 ± 0.1^{aAB}
phytotoxic >10[1]	MCa	1.7 ± 0.2^{abA}	0.4 ± 0.06^{bAB}	1.2 ± 0.3^{abB}	2.2 ± 0.2^{aAB}
	MP	1.8 ± 0.1^{abA}	1.1 ± 0.15^{bA}	3.2 ± 0.2^{aA}	3.4 ± 0.1^{aA}
	McC	0.9 ± 0.1^{aAB}	0.6 ± 0.1^{aAB}	1.7 ± 0.2^{aAB}	2.4 ± 0.4^{aAB}
K (g kg^{-1})	LC	5.6 ± 0.1^{bA}	–	24.9 ± 2.4^{aAB}	–
	LCa	6.4 ± 0.3^{bA}	12.4 ± 0.4^{abB}	23 ± 2^{aAB}	13.7 ± 1^{abAB}
deficient	LP	5.8 ± 0.3^{bA}	–	24.7 ± 3.2^{aAB}	–
normal 20–50[1]	MC	7.7 ± 0.8^{bA}	21 ± 2^{aAB}	30 ± 2^{aAB}	14 ± 1.5^{abAB}
phytotoxic	MCa	7.7 ± 0.25^{bA}	18 ± 2^{abAB}	32 ± 5^{aAB}	15 ± 1.4^{abAB}
	MP	7.2 ± 6.2^{bA}	17 ± 1.9^{aAB}	19 ± 2^{aB}	11 ± 0.9^{abB}
	McC	6.9 ± 0.7^{bA}	32 ± 7.5^{abA}	38.9 ± 1.4^{aA}	22.5 ± 4.0^{abA}
Ca (g kg^{-1})	LC	9.3 ± 0.99^{aAB}	–	13.5 ± 2.9^{aAB}	–
	LCa	5.9 ± 0.3^{abAB}	3.85 ± 0.3^{bA}	10.5 ± 0.8^{aAB}	3.45 ± 0.1^{bAB}
deficient	LP	9.6 ± 0.8^{aAB}	–	11 ± 1^{aAB}	–
normal 1–50[1]	MC	6.4 ± 0.65^{abAB}	3.8 ± 0.4^{bA}	6.9 ± 0.7^{aAB}	3.8 ± 0.4^{abAB}
phytotoxic	MCa	5.4 ± 0.6^{abB}	3.6 ± 0.4^{bA}	7.0 ± 0.4^{aAB}	3.4 ± 0.4^{bAB}
	MP	5.2 ± 0.4^{abB}	3.6 ± 0.6^{abA}	5.8 ± 0.7^{aB}	3.15 ± 0.2^{bB}
	McC	13.6 ± 0.7^{aA}	2.8 ± 0.3^{aA}	15.4 ± 1.9^{aA}	6.8 ± 0.4^{aA}
Mg (g kg^{-1})	LC	1.4 ± 0.1^{bAB}	–	3.3 ± 0.3^{aB}	–
	LCa	1.2 ± 0.1^{bB}	0.9 ± 0.1^{abA}	3.9 ± 0.2^{aAB}	1.6 ± 0.1^{bB}
deficient	LP	1.5 ± 0.2^{bAB}	–	3.4 ± 0.5^{aB}	–

Table 3 (continued)

Variable	Treatment	Organ			
		Belowground organs	Stems	Leaves	Seeds
normal 1.5–3.5[1]	MC	2.85 ± 0.5^{abA}	1.8 ± 0.2^{bA}	5.3 ± 0.2^{aA}	2.7 ± 0.3^{abA}
phytotoxic	MCa	1.7 ± 0.2^{bAB}	1.4 ± 0.2^{bA}	3.8 ± 0.15^{aAB}	2.2 ± 0.1^{abAB}
	MP	1.9 ± 0.2^{bAB}	1.5 ± 0.2^{bA}	3.9 ± 0.4^{aAB}	2.2 ± 0.1^{abAB}
	McC	1.3 ± 0.1^{abAB}	0.8 ± 0.1^{bA}	5.7 ± 0.4^{aAB}	2.8 ± 0.2^{abA}

– no material;1—Marschner (1995); 3—adapted from Pugh et al. (2002), 4—adapted from Levy et al. (1999), 5—adapted from Kabata-Pendias (2001), 6—Alkorta et al. (2004), 7—Mahler (2004), 8—Allen (1989), 9—Zhang et al. (2007), 10—Garcia-Salgado et al. (2012), 11—adapted from Lorestani et al. (2011), 12—adapted from Guleryuez et al. (2008), 13—Bose and Bhattacharyya (2008)

Concentration of microelements in the organs

The concentrations of Cu, Fe, Mn, and Ni were significantly affected by treatments, and the concentrations of Cu, Fe, Mn, Ni, and Zn differed between the individual organs (see Table 4 for details). The concentration of Cu ranged from 2.7 mg kg^{-1} in seeds in the LCa to 91 mg kg^{-1} in belowground organs in the LP treatment; the concentration of Fe ranged from 51.5 mg kg^{-1} in stems in the LCa treatment, to 5357 mg kg^{-1} in leaves in the MP treatment; the concentration of Mn ranged from 3.5 mg kg^{-1} in stems in the LCa treatment, to 228 mg kg^{-1} in belowground organs in the LC treatment; the concentration of Ni ranged from 0.5 mg kg^{-1} in stems in the LCa treatment, to 5.9 mg kg^{-1} in leaves in the MP treatment; and finally, the concentration of Zn ranged from 24 mg kg^{-1} in stems, to 83 mg kg^{-1} in belowground organs in the non-contaminated McC treatment.

Concentration of risk elements in the organs

The concentrations of As, Cd, Cr, Pb, and Zn were significantly affected by treatments and analysed plant organs (see Table 4 for details). The concentration of As ranged from 0.22 mg kg^{-1} in stems in the McC treatment to 189 mg kg^{-1} in leaves in the MCa treatment; the concentration of Cd ranged from 0.2 mg kg^{-1} in stems in the MCa treatment, to 29 mg kg^{-1} in belowground organs in the LC treatment; the concentration of Cr ranged from 0.07 mg kg^{-1} in seeds in the McC treatment, to 6.8 mg kg^{-1} in leaves in the MP treatment; the concentration of Pb ranged from 0.1 mg kg^{-1} in stems in the McC treatment to 235 mg kg^{-1} in belowground organs in the LC treatment; and finally, the concentration of Zn, in plants grown on contaminated soil, ranged from 30 mg kg^{-1} in stems in the MC and MCa treatments, to 1479 mg kg^{-1} in belowground organs in the LC treatment.

Bioaccumulation and translocation factors

Bioaccumulation factors for all elements were significantly affected by treatments (Table 5). In the non-contaminated McC treatment, the BF was above one only for Cd and Ni and in contaminated soils of LC and MC treatments, the BF was below one for all elements. Bioaccumulation factors for As, Cd, Cu, Mn, Ni, Pb, and Zn were affected by their level of soil contamination (As: $r=0.541$, $p=0.001$; Cd: $r=-0.360$, $p=0.040$; Cu: $r=-0.377$, $p=0.031$; Mn: $r=-0.587$, $p<0.01$; Ni: $r=-0.368$, $p=0.035$; Pb: $r=-0.457$, $p<0.01$ and Zn: $r=-0.376$, $p=0.031$). Liming (MCa treatment) and application of superphosphate (LP and MP treatments) did not affect the BF in contaminated soils.

Translocation factors for As, Cu, and Ni were significantly affected by treatments (Table 5). Liming (LCa and MCa treatments) and application of superphosphate (MP treatment) affected the TF in contaminated soils.

Result of PCA analysis

The first axis of the PCA analysis explained 35 %, the first two axes 56 % and the first four axes together, 82 % of the variability of all analysed data (Fig. 2). The first ordination axis divided individual pots into the Litavka group on the right side and Malín and Mšec groups on the left side of the diagram. This indicates an effect of soil properties on the availability of elements in soil and biomass production as well as on element accumulation.

Table 4 Continuation of Table 3

Variable	Treatment	Organ			
		Belowground organs	Stems	Leaves	Seeds
As (mg kg^{-1})	LC	50 ± 17^{aAB}	–	20 ± 8^{aABC}	–
	LCa	10.5 ± 4^{abAB}	1.7 ± 0.4^{bcAB}	23 ± 6^{aABC}	0.6 ± 0.4^{cdBC}
deficient -[5]	LP	59 ± 23^{aAB}	–	9.6 ± 2.6^{aC}	–
normal 1–1.7[5]	MC	155 ± 67^{abA}	12 ± 3^{bA}	127 ± 30^{aAB}	21 ± 7^{abA}
phytotoxic 5–20[5]	MCa	60 ± 28^{abAB}	7.6 ± 1.7^{bAB}	189 ± 84^{aAB}	6.6 ± 3.1^{bABC}
hyperaccumulation level 1000[10]	MP	82 ± 37^{abAB}	15.5 ± 4.7^{abA}	153 ± 30^{aAB}	17 ± 10^{bABC}
	McC	0.81 ± 0.17^{aB}	0.22 ± 0.06^{aB}	0.75 ± 0.22^{aC}	0.23 ± 0.05^{aBC}
Cd (mg kg^{-1})	LC	29 ± 4.5^{aA}	–	14 ± 2^{bA}	–
	LCa	6.8 ± 1.1^{aAB}	1.5 ± 0.3^{abA}	5.9 ± 0.9^{aABC}	1.2 ± 0.3^{bA}
deficient -[3]	LP	19 ± 3.8^{aA}	–	10.5 ± 1^{bAB}	–
normal 0.05–2[3]	MC	4.8 ± 1.6^{aAB}	0.3 ± 0.1^{bAB}	2.1 ± 0.4^{aC}	0.5 ± 0.1^{abAB}
phytotoxic 5–700[3]	MCa	2.1 ± 0.6^{aB}	0.2 ± 0.04^{bB}	1.9 ± 0.8^{abC}	0.2 ± 0.05^{bB}
hyperaccumulation level 100[6]	MP	2.8 ± 0.8^{aB}	0.5 ± 0.1^{abAB}	2.3 ± 0.4^{aABC}	0.5 ± 0.2^{bAB}
	McC	0.77 ± 0.12^{aBC}	0.45 ± 0.15^{aAB}	0.99 ± 0.24^{aC}	0.29 ± 0.15^{aAB}
Cr (mg kg^{-1})	LC	3.1 ± 1.1^{aA}	–	2.3 ± 0.6^{aAB}	–
	LCa	1.5 ± 0.8^{abA}	0.1 ± 0.03^{bB}	3.2 ± 0.8^{aAB}	0.2 ± 0.08^{bAB}
deficient -[5]	LP	3.0 ± 1.1^{aA}	–	1.3 ± 0.2^{aAB}	–
normal 0.1–0.5[5]	MC	6.2 ± 2.6^{abA}	0.6 ± 0.2^{bAB}	5.8 ± 1.3^{aAB}	1.6 ± 0.4^{abA}
phytotoxic 5–30[5]	MCa	2.4 ± 1.3^{abA}	0.3 ± 0.1^{bAB}	6.6 ± 2.6^{aAB}	0.7 ± 0.2^{abAB}
hyperaccumulation level 1000[9]	MP	3.2 ± 1.4^{abA}	0.9 ± 0.3^{bA}	6.8 ± 1.3^{aA}	1.4 ± 0.4^{abA}
	McC	0.82 ± 0.25^{aA}	0.14 ± 0.05^{abAB}	0.44 ± 0.09^{abB}	0.07 ± 0.01^{bB}
Cu (mg kg^{-1})	LC	78 ± 51^{aA}	–	9.5 ± 0.8^{bAB}	–
	LCa	13 ± 2^{aAB}	2.9 ± 0.9^{bA}	8.2 ± 1.05^{abAB}	2.7 ± 0.6^{bB}
deficient <1–5[3]	LP	91 ± 60^{aA}	–	11 ± 2^{bAB}	–
normal 4–15[12]	MC	25.5 ± 9^{aAB}	6.6 ± 3.6^{aA}	16 ± 2^{aAB}	11 ± 3^{aA}
phytotoxic 20–100[3]	MCa	12.5 ± 3^{aAB}	26 ± 15^{aA}	15 ± 5^{aAB}	4 ± 0.6^{aAB}
hyperaccumulation level 1000[6]	MP	24 ± 13^{aAB}	19 ± 10^{aA}	17 ± 2^{aA}	7 ± 0.9^{aA}
	McC	4.3 ± 0.5^{aB}	2.9 ± 0.15^{aA}	4.3 ± 0.3^{aB}	3.6 ± 0.2^{aAB}
Fe (mg kg^{-1})	LC	2463 ± 866^{aA}	–	1761 ± 703^{aAB}	–
	LCa	965 ± 473^{abA}	51.5 ± 13^{bB}	2092 ± 607^{aAB}	82 ± 34^{bB}
deficient <40[4]	LP	2337 ± 961^{aA}	–	795 ± 187^{aB}	–
normal 30–300[4]	MC	4547.5 ± 1954^{abA}	251.5 ± 97^{bAB}	4456 ± 1000^{aAB}	926 ± 258^{abA}
phytotoxic >500[4]	MCa	1857 ± 1011^{abA}	171 ± 57^{bAB}	5273 ± 2231^{aAB}	315 ± 124^{abAB}
hyperaccumulation level 10000[11]	MP	2426 ± 1058^{abA}	441 ± 144^{bA}	5357 ± 1051^{aA}	758 ± 387^{bAB}
	McC	432 ± 135^{aA}	62 ± 2^{abAB}	216 ± 89^{abB}	52 ± 4^{bAB}
Mn (mg kg^{-1})	LC	228 ± 96^{aA}	–	183 ± 82^{aA}	–
	LCa	116 ± 55^{abA}	3.5 ± 0.5^{bB}	207 ± 49^{aA}	25 ± 19^{abA}
deficient	LP	118 ± 61^{aA}	–	81 ± 23^{aA}	–
normal 40–200[7]	MC	71 ± 28^{abA}	4.65 ± 1.4^{bAB}	88 ± 33^{aA}	20 ± 5^{abA}
phytotoxic >356[13]	MCa	30 ± 15^{abA}	3.6 ± 0.9^{bAB}	77 ± 32^{aA}	10.5 ± 1.8^{abA}
hyperaccumulation level 10000[6]	MP	39 ± 16^{abA}	7.5 ± 1.7^{bAB}	83 ± 17^{aA}	21 ± 8.5^{abA}
	McC	56 ± 5.5^{abA}	17 ± 1^{bA}	199 ± 56^{aA}	64 ± 3^{abA}
Ni (mg kg^{-1})	LC	4.1 ± 1^{aA}	–	2.3 ± 0.6^{aAB}	–

Table 4 (continued)

Variable	Treatment	Organ			
		Belowground organs	Stems	Leaves	Seeds
	LCa	1.4 ± 0.4^{abA}	0.5 ± 0.05^{bA}	2 ± 0.4^{aAB}	0.6 ± 0.1^{bB}
deficient	LP	3.2 ± 0.8^{aA}	–	1.4 ± 0.2^{bB}	–
normal 0.5–5[8]	MC	5.3 ± 1.8^{aA}	0.7 ± 0.2^{bA}	4.8 ± 0.9^{aAB}	2.2 ± 0.7^{abA}
phytotoxic >5[8]	MCa	2.5 ± 0.85^{abA}	0.7 ± 0.1^{bA}	5.0 ± 1.8^{aAB}	1.2 ± 0.2^{abAB}
hyperaccumulation level 1000[6]	MP	3.3 ± 1.2^{abA}	1.0 ± 0.2^{bA}	5.9 ± 1^{aA}	1.8 ± 0.3^{abA}
	McC	1.0 ± 0.1^{aA}	0.6 ± 0.1^{aA}	1.2 ± 0.2^{aB}	1.2 ± 0.1^{aAB}
Pb (mg kg^{-1})	LC	235 ± 88^{aA}	–	142 ± 63^{aB}	–
	LCa	123 ± 54^{abAB}	2.9 ± 0.2^{bA}	166 ± 44^{aAB}	5.5 ± 3^{bA}
deficient -[3]	LP	113 ± 48^{aAB}	–	59 ± 19^{aABC}	–
normal 0.5–10[3]	MC	14 ± 5^{aAB}	1.2 ± 0.4^{aAB}	10.5 ± 2.5^{aC}	5 ± 1.9^{aA}
phytotoxic 30–300[3]	MCa	6 ± 2.5^{aB}	1.9 ± 0.6^{aAB}	13.6 ± 6.5^{aABC}	1.8 ± 0.3^{aA}
hyperaccumulation level 1000[6]	MP	7.6 ± 2.8^{aB}	2.2 ± 0.4^{aAB}	13 ± 2^{aABC}	6 ± 2.8^{aA}
	McC	1.6 ± 0.3^{aAB}	0.1 ± 0.1^{bB}	0.8 ± 0.3^{abC}	0.2 ± 0.1^{abA}
Zn (mg kg^{-1})	LC	1479 ± 360^{aA}	–	1260 ± 213^{aAB}	–
	LCa	329 ± 87^{abAB}	50 ± 7^{bA}	498 ± 105^{aABC}	55 ± 17^{bA}
deficient <10[3]	LP	809 ± 180^{aAB}	–	875 ± 66^{aB}	–
normal 10–150[3]	MC	231 ± 76^{aAB}	30 ± 7^{bA}	189 ± 39^{aC}	60 ± 9.5^{abA}
phytotoxic >100–500[5]	MCa	124 ± 46^{abB}	30 ± 8^{bA}	241 ± 100^{aABC}	32 ± 5^{bA}
hyperaccumulation level 10000[6]	MP	141 ± 48^{abB}	50 ± 9^{abA}	220 ± 40^{aABC}	54 ± 19^{bA}
	McC	83 ± 12^{aB}	24 ± 3^{aA}	74 ± 18^{aC}	28 ± 2^{aA}

In Litavka, in contrast to Malín soil, data for lime treatment (LCa) were clearly separated from all marks for control (LC) and superphosphate treatments (LP). This indicates a large effect of lime application on all the recorded data in Litavka soil and a minimal effect in Malín soil. In the majority of treatments, data for stems and seeds were grouped into the upper part of the diagram, indicating the lowest concentrations of elements in these organs, since the vectors for the majority of elements in the biomass grouped on the opposite site of the diagram.

The length and direction of the vectors relating to the individual elements indicate the association of elements with their respective treatments. For example, Zn concentration was the highest in belowground organs in the LC treatment, but the lowest in stems in the McC treatment. The concentration of Zn in plant biomass (Zn/B) was positively correlated with plant-available Zn (Zn/Ca) and also with acid-extractable Zn (Zn/AA) concentrations in the soil as indicated by an angle between the vectors for Zn/B and Zn/Ca or Zn/AA of less than 90°. The concentration of Zn in plant biomass was negatively correlated with biomass of organs (DM) as the angle between vectors for Zn/B and DM was greater than 90°. Two vectors did not positively correlate, if the angle between them is larger than 90°. A long vector for a particular variable indicates that it greatly affected the results of the analysis and the opposite was the case for a short vector. For example, there was no effect of soil and treatment on the concentrations of K and Mg in plant biomass, as vectors for these elements (K/B and Mg/B) were very short.

Discussion

Biomass production

Biomass production (i.e. total biomass of all organs) of *R. obtusifolius* was clearly negatively related to the concentrations of Cd, Ni, Pb, and Zn in its biomass, indicating their toxicity to plants. Very high concentrations of micro- and risk elements in plants (i.e. depending on the plant species, >5 mg As kg^{-1}, >5 mg Cd kg^{-1},

Table 5 Effect of treatment on bioaccumulation (BF) and translocation (TF) factors (mean ± SE). Treatment abbreviations: *LC* Litavka control soil without any additive, *LCa* Litavka soil with lime, *LP* Litavka soil with superphosphate, *MC* Malín control soil without any additive, *MCa* Malín soil with lime, *MP* Malín soil with superphosphate, and *McC* Mšec, non-contaminated control soil. Calculated by Kruskal–Wallis test, differences between treatments were not statistically significant ($^{n.s.}$) or were significant at 0.05 (*) and 0.01 (**) probability levels. According to the multiple comparisons of mean ranks, treatments with the same letter were not significantly different

Variable	Elements	Treatment						
		LC	LCa	LP	MC	MCa	MP	McC
BF	As *	0.06±0.02^{a}	0.04±0.01^{a}	0.03±0.01^{a}	0.1±0.03^{a}	0.1±0.04^{a}	0.1±0.04^{a}	0.05±0.01^{a}
	Cd **	0.3±0.04^{ab}	0.08±0.01^{b}	0.2±0.02^{ab}	0.1±0.02^{ab}	0.1±0.03^{b}	0.1±0.04^{ab}	2.9±0.7^{a}
	Cr **	0.04±0.01^{a}	0.04±0.01^{a}	0.03±0.005^{a}	0.09±0.02^{a}	0.09±0.02^{a}	0.1±0.03^{a}	0.01±0.002^{a}
	Cu *	0.2±0.01^{ab}	0.1±0.01^{b}	0.2±0.03^{ab}	0.2±0.02^{ab}	0.2±0.03^{ab}	0.2±0.04^{ab}	0.3±0.02^{a}
	Fe **	0.08±0.03^{ab}	0.06±0.02^{ab}	0.04±0.01^{ab}	0.2±0.05^{a}	0.2±0.05^{ab}	0.2±0.06^{a}	0.01±0.004^{b}
	Mn **	0.07±0.03^{ab}	0.05±0.01^{ab}	0.03±0.01^{b}	0.2±0.05^{ab}	0.1±0.03^{ab}	0.2±0.04^{ab}	0.3±0.08^{a}
	Ni **	0.01±0.01^{b}	0.4±0.1^{ab}	0.02±0.01^{ab}	0.4±0.1^{ab}	0.7±0.2^{a}	0.5±0.1^{ab}	1.5±0.6^{a}
	Pb **	0.04±0.02^{a}	0.03±0.01^{a}	0.02±0.01^{a}	0.08±0.02^{a}	0.08±0.02^{a}	0.1±0.03^{a}	0.01±0.005^{a}
	Zn **	0.2±0.03^{ab}	0.05±0.01^{b}	0.1±0.01^{ab}	0.1±0.03^{ab}	0.1±0.03^{ab}	0.2±0.04^{ab}	1.0±0.1^{a}
TF	As *	0.5±0.2^{ab}	2.7±0.8^{ab}	0.2±0.1^{b}	1.5±0.7^{ab}	4.5±1.7^{a}	3.9±2.6^{ab}	1.2±0.6^{ab}
	Cd $^{n.s.}$	0.5±0.1^{a}	1.0±0.2^{a}	0.5±0.04^{a}	0.4±0.1^{a}	0.9±0.3^{a}	1.3±0.5^{a}	1.2±0.1^{a}
	Cr $^{n.s.}$	0.9±0.4^{a}	3.3±1.1^{a}	0.4±0.1^{a}	1.5±0.6^{a}	3.4±1.0^{a}	5.1±3.6^{a}	0.7±0.3^{a}
	Cu *	0.1±0.1^{a}	0.7±0.1^{a}	0.2±0.1^{a}	0.9±0.2^{a}	1.3±0.3^{a}	1.4±0.4^{a}	1.0±0.2^{a}
	Fe $^{n.s.}$	0.8±0.3^{a}	5.2±1.8^{a}	0.4±0.1^{a}	2.0±1.1^{a}	3.0±1.1^{a}	5.1±3.6^{a}	0.7±0.4^{a}
	Mn $^{n.s.}$	0.8±0.3^{a}	2.5±0.9^{a}	0.7±0.2^{a}	1.9±0.9^{a}	2.6±0.9^{a}	3.6±2.2^{a}	3.5±0.7^{a}
	Ni *	0.5±0.02^{a}	1.9±0.1^{a}	0.4±0.03^{a}	1.4±0.4^{a}	2.4±0.8^{a}	1.9±0.7^{a}	1.2±0.2^{a}
	Pb $^{n.s.}$	1.0±0.4^{a}	2.3±0.6^{a}	0.8±0.3^{a}	3.0±1.5^{a}	2.0±0.4^{a}	4.8±3.8^{a}	0.6±0.3^{a}
	Zn $^{n.s.}$	0.8±0.2^{a}	1.9±0.4^{a}	1.2±0.2^{a}	0.9±0.2^{a}	1.6±0.4^{a}	2.7±1.0^{a}	0.9±0.1^{a}

BF—the ratio of total concentrations of elements in aboveground plant tissues (stems, leaves and seeds together) to pseudo-total concentrations of elements in soil ($BF = c_{tissues}/c_{soil}$)

TF—the ratio of total concentrations of elements in leaves to total concentrations of elements in belowground organs ($TF = c_{leaf}/c_{belowground\ organs}$)

>5 mg Cr kg^{-1}, >5 mg Ni kg^{-1}, >30 mg Pb kg^{-1} or >100 mg Zn kg^{-1}) can reduce biomass production, because micro- and risk elements can cause inhibition of cell elongation and division (Anton and Mathe-Gaspar 2005; Chen and Wong 2006; Barrutia et al. 2009). In LC and LP treatments, the toxicity of risk elements was high enough to inhibit the development of stems and generative organs. *R. obtusifolius* is thus, a species with a high sensitivity to metal(loid) toxicity.

The highest biomass production of *R. obtusifolius* in the McC treatment was probably connected with the lower micro- and risk elements concentrations (i.e. depending on the plant species, in range of 1–1.7 mg kg^{-1} for As, 0.05–2 mg kg^{-1} for Cd, 0.1–0.5 mg kg^{-1} for Cr, 0.5–5 mg kg^{-1} for Ni, 0.5–10 mg kg^{-1} for Pb or 10–150 mg kg^{-1} for Zn), but also to better N and K nutrition as shown by the N and K concentrations in leaves. In the Litavka soil, a greater biomass production in the LCa than the LC treatment was probably connected with the obvious trend that lime substantially reduced the mobility of micro- and risk elements, in particular of Cd, Mn, and Zn (Table 2). Similar results, i.e. an increased growth of several crops and weedy species on acid soils contaminated by Cd, Cu, Ni, Pb, and Zn after lime application has also been recorded by other authors (Chen and Wong 2006; Tlustoš et al. 2006; Alvarenga et al. 2008).

Concentrations of macroelements in plant organs

In the McC treatment, the highest concentrations of N, K, Ca, and Mg were recorded as expected, in leaves,

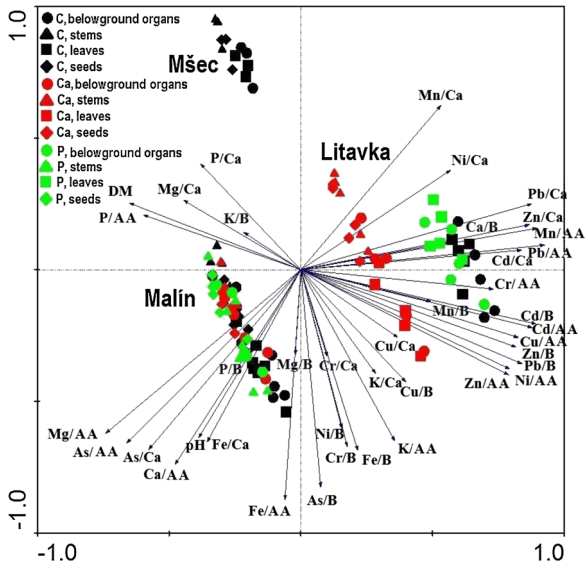

Fig. 2 Ordination diagram showing the results of PCA analysis with element concentrations in organs of *R. obtusifolius* plants grown on contaminated and non-contaminated soils. Soil abbreviations: *Litavka* slightly acidic contaminated soil, *Malín* alkaline contaminated soil, *Mšec* acidic non-contaminated soil. Treatment abbreviations: *C* control, *Ca* lime application, *P* superphosphate application; *C, belowground organs* concentrations of elements in belowground organs in control and etc. Element abbreviations:*/B* total concentrations of elements in plant biomass,*/Ca* plant-available concentrations of elements in soil (extracted by 0.01 mol L^{-1} $CaCl_2$) and*/AA* acid-extractable concentrations of elements in soil (extracted by 0.11 mol L^{-1} CH_3COOH). Other abbreviations: *DM* dry matter biomass per organ and *pH* mean soil pH

because leaves are the most metabolically active organs, with high nutrient requirements (López-Lefebre et al. 2001). The most surprising result was the highest Ca concentration in belowground organs, leaves and seeds in the McC treatment, despite the lowest soil Ca availability (Tables 1 and 2). Similarly, the Ca concentration in belowground organs and leaves in LC, LCa, and LP treatments was higher than in the MC, MCa, and MP treatments, despite a higher Ca availability in the MC, MCa, and MP treatments. The lowest availability of Ca in the McC treatment was reflected only by the lowest concentration of Ca in stems. The explanation for these discrepancies between Ca availability and Ca concentrations in belowground organs and leaves appears to be at least partly due to competition between Ca and Mg ions (Appenroth and Gabrys 2003). This is because of obvious trend that the highest concentrations of Mg in belowground organs were generally in the treatments where low concentrations of Ca were recorded. The highest concentration of P was recorded in seeds, due to the high P requirements for generative reproduction (Jiang et al. 2007; White and Veneklaas 2012). With the exception of the MP treatment, the P concentration in seeds was below the critical value of 3 g kg^{-1}, below which there is a decrease in the germination ability of *R. obtusifolius* or *R. crispus* (Hrdličková et al. 2011; Hejcman et al. 2012a). The lowest concentrations of N and P were recorded in stems and belowground organs, of Ca and Mg in stems and of K in belowground organs. A low concentration of macroelements in stems is connected with their low metabolic activity and with a high mobility of N, P, K, and Mg in plants and therefore, considerable translocation into plant apices (Anton and Mathe-Gaspar 2005; Gaweda 2009).

In the LCa treatment, a significant decrease in the concentrations of N and Ca in belowground organs after liming might be associated with a dilution effect caused by greater biomass production (Chen and Wong 2006; Tlustoš et al. 2006).

No effect of liming or superphosphate application on the distribution of K, Ca, and Mg in plant biomass was recorded in either acid- or alkaline-contaminated soils. Similarly, no effect of liming on the Mg concentration in plant tissues of other weedy species was found by Alvarenga et al. (2008).

Concentrations of microelements in plant organs

In the McC treatment, the highest concentrations of Fe and Zn were recorded in belowground organs; that of Cu in belowground organs and leaves; of Mn in leaves; the lowest concentrations of Cu, Mn, and Zn in stems; and of Fe in seeds. Similar concentrations of Ni were recorded in all organs. Variability of all microelements in different organs might be due to compartmentalisation and translocation in the vascular system (Bose and Bhattacharyya 2008; Hansch and Mendel 2009). Similar results, i.e., concentrations of Cu, Fe, and Ni in the order belowground organs > leaves > stems is consistent with results for *R. acetosa*, but inconsistent for those for Cu, Fe, and Zn in *R. dentatus* when both were grown in non-contaminated soils (Barman et al. 2000; Gaweda 2009). A different distribution of Mn was recorded, with the order leaves > seeds > belowground organs > stems, which clearly separated the distribution of Mn in *R. obtusifolius* from that of other microelements. This result is inconsistent with the distribution of Mn in *R. acetosa* (belowground organs > leaves > stems,

Gaweda 2009), which shows a difference in Mn distribution within *Rumex* species.

In LC and MC treatments, a tendency for a restricted transport of microelements from belowground organs into leaves in comparison to the McC treatment was recorded. This was probably connected with protection against excessive concentrations of microelements in aboveground organs (i.e. depending on the plant species as well as organ, >20 mg Cu kg^{-1}, >500 mg Fe kg^{-1}, >356 mg Mn kg^{-1}, >5 mg Ni kg^{-1} or >100 mg Zn kg^{-1}, Hansch and Mendel 2009).

In LCa and MCa treatments, a tendency for increased transport of Cu, Fe, Mn, and Ni from belowground organs into leaves in comparison to LC and MC treatments was recorded, as also demonstrated by higher TFs. We speculate that changes in the distribution pattern of microelements are connected to the presence of organic acids (mainly oxalate) for the formation of stable complexes, similar to the internal defence mechanism of oxalate plants against excess Ca (Tolra et al. 2005; Miyagi et al. 2013). Therefore, we can speculate that micro- and risk elements are precipitated with oxalate in roots in contaminated control soils. On the other hand, in contaminated soils with lime, oxalate is precipitated with Ca and thus is not available for micro- and risk elements that can easy transport to leaves. In the MP treatment, an increased transport of all microelements from belowground organs into leaves in comparison to the MC treatment was recorded probably because of the sufficient amount of Ca available from superphosphate as well as from soil solution precipitated oxalates as Ca-oxalate and thus available microelements can be easily transported to leaves. Changes in the translocation of microelements in plants after liming and superphosphate application require further research that focuses on differences between oxalate and non-oxalate plants.

Concentrations of risk elements in plant organs

In the McC treatment, the highest concentrations of risk elements (As, Cd, Cr, and Pb) were recorded in belowground organs or leaves and the lowest were recorded in stems and seeds. This was connected with low concentrations of risk elements in reproductive organs and with their lower metabolic activity in stems (Anton and Mathe-Gaspar 2005; Bose et al. 2008; Gaweda 2009).

In the LC and MC treatments, there was a higher transport of risk elements from stems into seeds and restricted transport from belowground organs into leaves in comparison to the McC treatment. The clear tendency for the accumulation of risk elements in belowground organs was connected with the exclusion strategy of *R. obtusifolius* and the function of roots as a barrier that limits the translocation of risk elements from the soil to the aboveground organs in soils contaminated by risk elements (Gaweda 2009; Zhang et al. 2010).

In the LCa and MCa treatments, there was a tendency for a greater transport of As, Cd, Cr, Pb, and Zn from belowground organs into leaves in comparison to the LC and MC treatments, in the most cases demonstrated also by higher TFs.

The results for Cd, Pb, and Zn transport were inconsistent with those for *Triticum aestivum* published by Tlustoš et al. (2006)—i.e. decreasing shoots/roots ratio after lime application in comparison to *R. obtusifolius*. As described above, we presume that oxalate plants possess an internal defence mechanism against risk elements, such as forming Ca-oxalate (Miyagi et al. 2013), because risk elements (mainly divalent Cd, Pb, and Zn) compete with divalent Ca for sites to form complexes with oxalate. There was a tendency for a higher transfer of Cd and Zn from belowground organs into leaves in the LP and MP treatments, in comparison to the LC and MC treatments, partly also demonstrated by higher TFs. Similar results were obtained for As and Pb, but only in alkaline-contaminated soil. This was demonstrated also by higher TFs. The results for Cd and Zn transport are inconsistent with observations for *Zea mays* and *Brassica parachinensis* (Jiang et al. 2007; Qiu et al. 2011). Therefore, we speculate that differences in distribution can be connected to the presence of oxalate (available for the formation of less toxic complexes as well as internal defence mechanism) in *R. obtusifolius*. Using lime and superphosphate application increased the in vivo mobility of As, Cd, Cr, Pb, and Zn into leaves of *R. obtusifolius*. For this reason, differences in the translocation of risk elements after liming and superphosphate application deserve closer examination, with a focus on the differences between oxalate and non-oxalate plants.

Bioaccumulation and translocation factors

In the non-contaminated McC treatment, the BF of micro- and risk elements for *R. obtusifolius* ranged from 0.01 to 2.9 and in the LC and MC treatments, ranged

from 0.01 to 0.4. A decrease in the BF with increasing pseudo-total concentrations of elements (Cd, Zn, Pb, Ni, Mn, and Cu) in soils was consistent with results of Cd and Zn published by Zhao et al. (2003). Therefore, we do not recommend the use of *R. obtusifolius* for phytoextraction in heavily contaminated soils, but it might be suitable for moderately contaminated soils, as is *R. acetosa* (Gaweda 2009). In addition, because of the sensitivity of *R. obtusifolius* to risk elements, it can be used for the identification of contaminated soils by field vegetation mapping, i.e. according to the symptoms of risk elements toxicity visible on aboveground organs in different phenological stages (Hejcman et al. 2012b). Liming tended to decrease the BF of micro- and risk elements in the contaminated Litavka soil, due to the reduction in element availability in soil and subsequently in plants. In the LP, MCa and MP treatments, no effect on the BF of micro- and risk elements was observed, because there was no reduction of element availability in the soil.

In the LC treatment, the TFs for risk elements (As, Cd, Pb, and Zn) ranged from 0.5 to 1.0, indicating an exclusion strategy by *R. obtusifolius*. In the MC treatment, the TFs for risk elements were higher, confirming that the ability to exclude risk elements was affected by their availability in the soil. The classification of *R. obtusifolius* as a metal-excluder with restricted risk element transfer to aboveground organs is consistent with observations for *R. acetosa* (Barrutia et al. 2009; Gaweda 2009). It appears that there is consistency in the physiological responses of different *Rumex* species to the availability of risk elements in the soil, but this requires further research. Plants with the ability to accumulate risk elements can be used to phytoremediate contaminated soils (Baker 1981). Therefore, the TF is only relevant for elements that exceed background concentrations in comparison to those of non-contaminated soils (see Table 1). In the LCa and MCa treatments, the TFs for As, Cd, Pb, and Zn ranged from 0.9 to 4.5, which is characteristic for indicators or accumulators. Liming substantially increased the translocation of risk elements from belowground organs into leaves. A similar result was observed for superphosphate application, but only in alkaline-contaminated soil. We conclude that the identification of plants for the phytoremediation of contaminated soils must proceed with caution, because TF values depend on the chemical properties of the soil.

Conclusions

The ionome, i.e., the elemental composition of different organs is greatly affected by soil chemical properties. Soil chemical properties affect not only the concentrations of individual elements in individual organs, but also their distribution between plant organs. Variability in the concentrations of micro- and risk elements is much greater than variability in the concentrations of macroelements, especially on metal(loid)-contaminated soils. Liming of contaminated soils as well as superphosphate application can modify the distribution pattern of elements and can increase the translocation of micro- and risk elements from belowground organs to leaves.

The oxalate plant, *R. obtusifolius*, is sensitive to Cd, Ni, Pb, and Zn toxicity, as its biomass production is reduced due to their availability in the soil and consequently due to reduction of their high concentrations in plant organs. The restricted translocation of micro- and risk elements from belowground organs to leaves in *R. obtusifolius* is consistent with this species being an As-, Cd-, Pb-, and Zn-excluder and not suitable for phytoremediation of heavily contaminated soils. However, sensitivity to risk elements can be used for identification of metal(loid)-contaminated soils by field vegetation mapping.

Acknowledgments The finalisation of this manuscript was supported by projects NAZV QJ 1210211 and by the project CIGA 20124205.

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