



The phytoextraction power of *Cichorium intybus* L. on metal-contaminated soil: Focus on time- and cultivar-depending accumulation and distribution of cadmium, lead and zinc

Théo Guérin, Alina Ghinet, Christophe Waterlot

► To cite this version:

Théo Guérin, Alina Ghinet, Christophe Waterlot. The phytoextraction power of *Cichorium intybus* L. on metal-contaminated soil: Focus on time- and cultivar-depending accumulation and distribution of cadmium, lead and zinc. *Chemosphere*, 2022, 287, pp.132122. 10.1016/j.chemosphere.2021.132122 . hal-03771356

HAL Id: hal-03771356

<https://hal.science/hal-03771356>

Submitted on 16 Oct 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial 4.0 International License

Title: The phytoextraction power of *Cichorium intybus* L. on metal-contaminated soil: focus on time- and cultivar-depending accumulation and distribution of cadmium, lead and zinc.

Authors: Théo Guérin^{1,2}, Alina Ghinet^{2,3,4}, Christophe Waterlot^{1*}

Affiliations:

¹ Univ. Lille, Institut Mines-Télécom, Univ. Artois, JUNIA, ULR 4515 – LGCgE, Laboratoire de Génie Civil et géo-Environnement, F-59000 Lille, France

² JUNIA, Health and Environment, Laboratory of Sustainable Chemistry and Health, F-59000 Lille, France

³ Univ. Lille, Inserm, CHU Lille, Institut Pasteur Lille, U1167 - RID-AGE - Facteurs de risque et déterminants moléculaires des maladies liées au vieillissement, F-59000 Lille, France

⁴ Faculty of Chemistry, Department of Organic Chemistry, ‘Alexandru Ioan Cuza’ University of Iasi, Bd. Carol I, nr. 11, 700506, Iasi, Romania

*Corresponding author: Christophe Waterlot (christophe.waterlot@junia.com)

Abstract

This study is focused on the evaluation of the accumulation of Cd, Pb and Zn in five cultivars of *Cichorium intybus* L. (chicory) which were produced on contaminated agricultural soil. Over a growth period of 211 days, the roots and leaves were collected in four stages and then analyzed by flame atomic absorption spectrometry to measure the concentration and distribution of the target metals in these two chicory organs considering the weather and the nature of the cultivar. For all cultivars, sharp decreases of Pb (from 165 mg kg⁻¹ to 3 mg kg⁻¹), Cd (from 11 mg kg⁻¹ to 5 mg kg⁻¹) and Zn concentrations (from 157 mg kg⁻¹ to 40 mg kg⁻¹) in the roots were highlighted over time. The data collected enabled the calculation of the variation of the bioconcentration factor, the biological absorption coefficient and the translocation factor for Cd, Pb and Zn. These parameters were then correlated with the distribution of the fresh biomass of leaves and roots and several indicators such as chlorophyll content, flavonols, anthocyanin and nitrogen balance index were measured. The study concludes with the discussion on the ability of chicory to clean up contaminated agricultural soil. The current investigation has shown: i) a translocation of Cd (and Zn to a lesser extent) from the roots to the leaves; ii) an increase in the level of anthocyanins with the increase of the metal trace elements concentration in the leaf, while the content of chlorophyll and the nitrogen balance index decrease, which could be linked to the phenomenon of senescence; iii) an ability of the chicory to reduce the bioavailable pool of the three metal trace elements studied, in particular for Cd.

Key words: Metal; Chicory; Bioavailability; Plant pigments; Remediation factor

1. Introduction

Soil pollution by **metal(loid)s** remains a major environmental concern in the whole world. These contaminants accumulate quickly in living organisms (fauna and flora) and often reach toxic concentrations that affect the soil functioning, the ecosystem, and the human health due to their long-term persistence in the environment. Human activities (*e.g.* mining and smelting of metal ores) are the main sources that contributed to increase the environmental anthropogenic pollution by **metal(loid)s** (Alloway, 1995, 2013). The most famous contaminated area in the Hauts-de-France region (France) is located in the former coal mining and ore processing sites (Metaleurop Nord and Nyrstar). For more than a century, the main pollutants released by these two metallurgical plants were lead (Pb), cadmium (Cd), and zinc (Zn), **defined as metal trace elements (MTE) in the current study**. The total concentration of these MTE and their bioavailability **in the surrounding agricultural soils** is sometimes problematic since a high proportion of the agricultural crops for foodstuffs are not conform with the European legislation (Douay et al., 2013; Pruvot et al., 2006).

Many *ex-situ* and *in-situ* methods have been developed to manage polluted soils (Gong et al., 2018) (*e.g.* encapsulation, soil washing, vitrification...). While these methods allow effective soil treatment, these technologies are often costly, time consuming and unsuitable for managing the soil of large polluted sites (Liu et al., 2018). Soil phytoremediation is a much more viable alternative because of its ease of implementation and the low costs it implies. This technology refers to the use of plants and amendments and/or microorganisms that can be added to improve the soil quality through several processes such as the degradation of organic matter, the degradation of pollutants as well as their extraction or stabilization (Ali et al., 2013). Among these processes, phytoaccumulation consists in **metal(loid)s** accumulation in the plant organs after their extraction from the polluted soils. **Some of them namely nickel (Ni), copper (Cu), manganese (Mn) and Zn are considered as essential for the growth of plants and so, they can accumulate them** (Bhargava et al., 2012). Some others like Cd, Pb, silver (Ag), arsenic (As), antimony (Sb) or mercury (Hg) are not essential and can cause phytotoxic effects (Finnegan and Weihua, 2012; Sheoran et al., 2016; Shtangeeva et al., 2011). However, metallophytes are able to concentrate some of these **metal(loid)s** from contaminated soils within their organs without necessarily significantly alter their growth and development (Rossini-Oliva et al.,

2018). Therefore, organ of these plants can be used through many applications to develop the bio economy and so ingrate the circular economy concept (Deyris et al., 2019; Hazotte et al., 2017; Gomes, 2012; Guérin et al., 2021; Hechelski et al., 2018; Janus et al., 2015; Oustrière et al., 2019; Waterlot et al., 2019).

Cichorium intybus L. (chicory) adapts to wide temperature ranges, different types of soils and is able to grow along the roadside, in industrial sites, urban parks and wasteland, even under high level of contamination (Askoy, 2008). Several studies have shown that chicory is capable, to some extent, of accumulating MTE (Cd, Pb and Zn) when produced on moderately contaminated soil (Mandzhieva et al., 2016; Sekara et al., 2005; Zeng et al., 2019). Moreover, Zeng et al. (2019) showed that translocation of metals to shoots depended on the nature of metals and on seedling and mature periods.

The purpose of this work is (i) to assess the dynamic of the MTE accumulation (Pb, Cd and Zn) by the roots and shoots of various cultivars of chicory produced on severely contaminated agricultural soils sampled near the former Metaleurop Nord plant, (ii) to examine the variation of several plant pigments (chlorophyll, anthocyanins, flavonols and nitrogen balance index) and (iii) to evaluate the ability of chicory in the remediation of contaminated soils by Cd, Pb and Zn.

2. Materials and Methods

2.1. Soil sampling, experimental set-up, and plants sampling

Contaminated soil (300 kg) was sampled from the ploughed layer (0-25 cm) of an agricultural plot (latitude: 50° 26' 15.9" N; longitude: 3° 01' 04.9" E) located not far away from the former Metaleurop Nord plant (France) in November 2018. This soil is a clay sandy loam dominated by silt and it is characterized by the following parameters: pH=7.7, Total N: 1200 mg kg⁻¹; Total C: 18.2 g kg⁻¹; C/N: 15.2; P₂O₅: 160 mg kg⁻¹; Organic matter: 45 g kg⁻¹; CEC: 15 cmol⁺ kg⁻¹ (Hechelski et al., 2020; Nsanganwimana et al., 2015). After homogenization, five containers (outside length: 40 cm; outside width: 30 cm; outside height: 25 cm) were filled with 20 kg of air-dried contaminated soil. The soil moisture experimental device was maintained at 40% field capacity with tap water in greenhouse at 20-25 °C. In each of containers, 7 seeds of chicory were sown in 2 rows. Five cultivars, providing by Florimond Desprez company (<https://www.florimond-desprez.com/rep/en/nl/groupe-florimond->

desprez-2.html), were considered (C1, C2, C3, C4 and C5) in the current study. Chicories were samples after 74, 105, 156 and 211 days after sowing, taking one plant out of every two times. Shoots and roots of chicory were then separated, crushed, and dried at 105 °C, then stored in acid-washed plastic bottles until digestion.

2.2. Digestion of plant and soil

2.2.1. Soil digestion and extractable-MTE in soils

300 mg of soil were digested by a mixture of 37% HCl (4.5 mL) and 70% HNO₃ (1.5 mL). The suspension was heated at 120 °C for 90 min in a hot-block digester (HotBlock™ Environmental Express®SC100, Charleston, USA). Ultra-pure water was then added until reaching 25 mL and the mixture was filtered over an acetate Millipore membrane (0.45 µm porosity). The resulting solutions were stored in acid-washed plastic bottles at 4 °C prior to analysis. Calcium chloride (0.01 M) and a mixture of Low Molecular Weight Organic Acids (LMWOAs) were used to evaluate the labile pool MTE in soils, the bioavailable MTE in short and long run (Waterlot et al., 2017; Waterlot and Hechelski, 2019).

2.2.2. Plant digestion

300 mg of dry plant sample (roots or leaves) were added to 5 mL of 70% HNO₃. The resulting suspension was heated at 95 °C for 90 min using the hot-block digester. After heating, 5 mL of H₂O₂ (30%) were slowly added in the cooled solution and the mixture was heated at 90±5 °C for 180 min. Ultra-pure water was then added until reaching a total volume of 25 mL, the mixture was filtered over an acetate Millipore membrane (0.45 µm porosity) and stored at 4 °C prior to analysis.

2.2.3. Chemical and statistical analyses

The concentration of MTE (Pb, Cd and Zn) in soil and plant digests were determined by flame or electrothermal atomic absorption spectrometry (AA-6800, Shimadzu, Tokyo, Japan) following the recommendations described in the literature (Waterlot et Douay, 2009; Savio et al., 2010). The limit of detection was 2.1 µg Cd L⁻¹, 41 µg Pb L⁻¹, 1.5 µg Zn L⁻¹ using flame as atomizer and 0.06 µg Pb L⁻¹ using graphite furnace as atomizer (Waterlot et al., 2013; Waterlot and Hechelski, 2019). INCTL-OBTL-5 (Virginia tobacco leaves *n*=3) was used as reference material for the quality control. The

digestion and analysis procedures yielded good average recovery values of 90-108 %, depending on MTE (Obtained concentrations: [Cd] = 2.36±0.12 mg kg⁻¹; [Pb] = 2.11±0.21 mg kg⁻¹; [Zn] = 56.2±2.2 mg kg⁻¹; Certified concentrations: [Cd] = 2.64±0.14 mg kg⁻¹; [Pb] = 2.01±0.31 mg kg⁻¹; [Zn] = 52.4±1.8 mg kg⁻¹).

Chlorophyll content (µg cm⁻²), Flavonols Index (relative units) and Anthocyanins Index (relative units) and were measured using a plant pigment content meter (Dualex® Scientific+, Force-A, Orsay, France) on green leaves at the base of the rosette. Moreover, the Dualex device provided a Nitrogen Balance Index (NBI) as reported in Rusinowski et al. (2019). Data collected were compared with those obtained from chicories grown in the same conditions on a control unpolluted soil considered as a reference soil (R). From the data set (n₁ = 120; n₁ = 6n with n=20), analyses were performed using the software program XLSTAT 2021 (Addinsoft, Paris, France). Significant differences between reference and/or cultivars were determined using a one-way ANOVA. Normality and homoscedasticity of residuals were met for all tests. When significant differences occurred between reference and/or cultivars, multiple comparisons of mean values were made using post-hoc Tukey HSD tests. Differences were considered statistically significant at p<0.05.

2.3. Indices calculation

2.3.1. Bioconcentration factor (BCF)

The BCF is the ability of plants to accumulate MTE with respect to their concentration in soils (Wang et al., 2019). In the current study, the BCF is the ratio of MTE concentration in the roots to those in the soil substrate (Eq. 1; Mirzaei et al., 2021):

$$BCF = \frac{C_{MTERP}}{C_{MTEs}} \quad (1)$$

where C_{MTERP} is the MTE concentration in the root of plants and C_{MTEs} is the MTE concentration of in soils.

2.3.2. Biological absorption coefficient (BAC)

The BAC is related to the bioaccumulation of MTE in the aerial part of plants. This index is defined as the ratio between the MTE concentrations in the shoots and the soil substrate (Eq. 2, Zehra et al., 2020).

$$BAC = \frac{C_{MTE}^{SP}}{C_{MTE}^S} \quad (2)$$

where C_{MTE}^{SP} is the MTE concentration in the shoot of plants and C_{MTE}^S is the MTE concentration in soils.

2.3.3. Translocation factor (TF)

The TF indicate the ability of plants to translocate MTE from roots to leaves (Zehra et al., 2020) and is defined as follows (eq 3):

$$TF = \frac{C_{MTE}^L}{C_{MTE}^R} \quad (\text{eq 3})$$

where C_{MTE}^L is the concentration of MTE in leaves and C_{MTE}^R is the concentration of MTE in roots.

3. Results and discussion

3.1. Metal concentration in soil-chicory system

The concentrations of Pb, Cd and Zn in the studied agricultural soil were $757.1 \pm 35.4 \text{ mg kg}^{-1}$, $10.7 \pm 0.5 \text{ mg kg}^{-1}$, $857.4 \pm 74.0 \text{ mg kg}^{-1}$ ($n = 9$), respectively. On average, these concentrations are 20-fold, 25-fold and 12-fold higher than those in the loessic reference soils (Sterckeman et al., 2002). Compared to the ploughed horizons of cultivated plots around Noyelles-Godault plants, Pb and Zn concentrations in the studied agricultural soil were the highest (45% and 33%, respectively) whereas Cd was quite similar (10.7 vs 9.8 ; Sterckeman et al., 2002). The 0.01M CaCl_2 -extractable concentrations were $0.04 \pm 0.01 \text{ mg Pb kg}^{-1}$, $0.28 \pm 0.07 \text{ mg Cd kg}^{-1}$ and $0.56 \pm 0.11 \text{ mg Zn kg}^{-1}$ ($n = 9$). The extractable-MTE concentrations measured in the mixture of LMWOAs were $5.34 \pm 1.07 \text{ mg Pb kg}^{-1}$, $1.83 \pm 0.12 \text{ mg Cd kg}^{-1}$ and $172 \pm 13 \text{ mg Zn kg}^{-1}$ ($n = 9$). These results highlight the poor labile pool of these MTE. The bioavailable fraction of MTE in short run was below 0.1%. In long run, it was 0.7% for Pb, 17% for Cd and 20% for Zn. This means that a potential decrease of the total Cd and Zn will be done, but obviously it will take time.

[Figure 1](#) shows the mean of MTE concentrations in the roots and shoot of chicory over time. Pb-concentrations in the roots of chicory at the first sampling (74 days) ranged from 101 to 165 mg.kg⁻¹, depending on cultivars. These concentrations were 10-fold higher than those measured in the shoots of cultivars C1, C2 and C5 and 6-fold higher for C4. From the second sampling up to the sampling at the end of the experiment, the concentration measured in the roots of chicory drastically decreased (from 60% to 85%) to reach a plateau, ranging from 3.21 to 9.07 mg Pb kg⁻¹ for the two last samplings, which depends on cultivars. In the same time, slight modifications in the Pb concentrations measured in the shoots of chicory may be noted but without any relations with the decrease of Pb concentrations in the roots. These results are not surprising knowing that Pb is a non-phytoavailable MTE and suggests a dilution of Pb in the roots over the growing period. **Indeed, the ratio of roots biomass between the second and the first samplings (105 and 74 days, respectively) is approximately 14.** However, these results are somewhat at odds with those described in Zeng et al. (2019). The mean Pb concentrations measured by the authors in the roots and shoots of chicory were around 50 and 25 mg kg⁻¹, although the Pb concentration in their soil were 10-fold lower at least than in the current study. This result means that the labile pool of Pb is a crucial parameter to consider. The extractable-MTE concentration using the mixture of LMWOAs and 0.01 M CaCl₂ represented 1% and 0.01% of the total Pb concentration, respectively, as reported in previous studies (Hechelski et al., 2020; Nsanganwimana et al., 2015). These data correlated well with the fact that the Pb concentrations in both studied chicory' organs are low despite the high total Pb concentrations in the soil substrate.

Variations in Cd and Zn concentrations in roots and shoots of chicory over time are presented in [Figures 1B and 1C](#). The concentrations of Cd and Zn in the roots of chicory decreased over time (from 50 to 60% and 64 to 78%, respectively) and slightly increased at the fourth sampling (211 days). In contrast, concentration of the two metals increased over the growth period, from 25 to 147% for Cd and from 5.5 to 43% for Zn. As shown on [Figures 1B and 1C](#), the Cd and Zn accumulations in plant organs at the end of the experiments were higher in shoots than in roots whatever the cultivars are. 211 days after sowing, the Cd and Zn concentrations in the shoots of chicory were up to 50 mg kg⁻¹ DW and 172 mg kg⁻¹ DW, respectively. These concentrations were, respectively, 7-fold and 3.4-fold higher than those in chicory' roots. These results correlated well with those obtained for Zn by Zeng et al.

(2019) but are quite different for Cd. No significant difference was highlighted by the authors between the Cd concentrations measured in shoots and in roots in seedling and mature periods. It has been reported in previous studies that the MTE accumulation capacities are affected by interspecific differences, environment conditions (Grant et al. 2008; Shi and Cai 2009) and the available Cd and the total Cd concentration, which was 8-fold higher in the current study than in Zeng et al. (2019).

3.2. Bioaccumulation factor, biological absorption coefficient and translocation factor

MTE-accumulation, MTE-translocation, and MTE-distribution in the upper and underground parts of plants were studied by means of three indices named bioconcentration factor (BCF: root-to-soil metal concentration ratio), biological adsorption coefficient (BAC: plant leaves-to-soil metal concentration ratio) and translocation factor (TF: leaves-to-root metal concentration ratio). The utilization of these indexes was recommended to define the ability of plant models in the phytoextraction and phytostabilization of MTE (Zhou et al., 2020). According to Alagić et al. (2015), a value of BCF greater than 1 ($BCF > 1$) indicates a good bioaccumulation of MTE in the roots of plants and if these plants have BAC values greater than 1 ($BAC > 1$), they are a suitable option for bioaccumulation of MTE in their shoots. Moreover, if $TF > 1$, the good ability of plants to translocate MTE from their roots to their aboveground parts make them as suitable for phytoextraction purpose (Vamelari et al., 2010). In contrast, for $BCF > 1$ and $TF < 1$, the potential of plants to be MTE-tolerant is great and so, they have potential for introducing as phytostabilization purpose (Vamelari et al., 2010). The three parameters were reported in Table 1 for each sample. The Pb-BCF values were low, ranging from 0.13 to 0.22 at the first sampling, and then, decreased sharply to reach 0.01 from the third sampling up to the last one. This trend was not cultivar-dependent. As shown in Table 1, the Pb-BAC values are very low over time (ranging from 0.01 to 0.03) whatever the cultivar meaning that chicories should not be suitable for Pb-phytoaccumulation. However, depending on time, the Pb-TF values were higher than 1 and were cultivar-dependent (from 1.44 to 3.53 at 156 days and from 0.86 to 1.34 at 211 days). These trends correlated well with the fact that the Pb-concentration remained quite constant from the second harvest to the end of the experiment. Caution should be taken when interpreting the results related to Pb-BAC and Pb-TF. Thus, we do not want to conclude on the

capacity of chicory to phytostabilize and phytoextract Pb. However, it is worth mentioning that chicories were described in other studies as Pb-phytoextractor (Vamelari et al., 2010) and Pb-accumulator (Baker et al., 1981).

As reported in Sêkara et al. (2005), Mandzhieva et al. (2016) and Zeng et al. (2019), the data for Cd distribution suggest that chicories are characteristic of good accumulators since BAC and TF were higher than 1. These two indicators increased over the growth period to reach values higher than 4 and 8, respectively corroborating the decrease of the BCF values over the same period. Unfortunately, due to the size of containers, the growth of plants was limited between 156 days and 211 days, potentially leading to a bias in the data collected. This result is even more pertinent as 60% of the total Cd-concentration was found in the acido-soluble fraction of the fourth-step sequential extraction procedure (Pelfrêne et al., 2015). Thus, by removing the shoots of chicory from the field, this plant should be able to be used for the removal of the bioavailable pool of Cd from the soil. Although Cd and Zn have similar chemical properties, the Zn-BAC and Zn-BCF were lower than 1 from the first harvest to the last one. However, Zn-BAC decreases whereas Zn-BCF increases over the growth period meaning that Zn-TF increases. As shown in Table 1, this indicator increases in time up to 156 days and was higher than 3 at 211 days meaning that chicories are able to extract Zn from soil. Recently, Wolf and Paulino (2020) pointed out the potential toxicity effects of Zn when chicories were produced on Zn-contaminated soils. In fact, the authors spiked their soils with zinc sulfate to obtain concentrations of 1200 mg Zn kg⁻¹ and 2400 mg Zn kg⁻¹. They highlighted a decrease of leaf biomasses 45 days after seedling and chlorosis effects. In the current study, no visual symptoms were noticed.

3.3. Plant uptake and foresight

The total harvested-plant biomass was expressed as leaf and root fresh-biomass distribution (Figure 2). As reported on it, the absolute mass distribution of these two plant-parts depends on cultivars. Leaf and root biomasses were in the same order of magnitude for the first two cultivars whereas they were different for the last three cultivars. For those, biomasses of roots ranged from 58.6% to 59.5% and so, were higher than in leaves. Based on the total mass of sampled roots and

leaves, exported metals from the soil to the chicories were calculated and presented in Table 2. The total metal uptake in leaves and roots were slightly dependent on cultivars and were highly metal-dependent. The ascending order of metals mass in the leaves of chicories was $Zn > Cd > Pb$ whereas it was $Zn > Pb > Cd$ in roots. Whatever the results, the data highlighted that if chicories are produced on contaminated soils, collections of leaf are required to avoid the recycling of leaf-bound MTE before the root harvest. As shown in Figure 3, chlorophyll contents in the reference (R), and the cultivars C3 and C5 were statistically the same. This result corroborates with the fact that the exported MTE by these two cultivars were the lowest. On the contrary, statistical differences were highlighted between the chlorophyll contents in R and the cultivars C1, C2 and C4, for which the exported MTE were the greatest. This means that higher the exported MTE is, lower the concentration of chlorophyll in the leaves of chicory is. Indeed, among the survival mechanisms of plants are MTE-translocation to leaves and decreasing chlorophyll (Uzu et al., 2010). Moreover, the decrease of chlorophyll may be linked to the senescence (Wojciechowska et al., 2018).

Although there is no statistical difference in the concentration of flavonols between the reference and the five cultivars studied, it is worth mentioning the relationship between the results obtained with chlorophyll and anthocyanins. No statistical difference was highlighted between anthocyanins in R and C3 and C5, whereas anthocyanin concentrations in R were statistically different from those in C1, C2 and C4. As shown in Figure 3, concentrations of anthocyanin were the highest in these three cultivars. This corroborates with previous studies in which it was well established that one of the role of anthocyanin is the protection of plants from several abiotic and biotic stress (Chalker-Scott, 1999; Ahmed et al., 2014), which may provide them a better adaptation to contamination level. Acting as antioxidant, anthocyanins are able to bind with MTE reducing consequently their toxicity (Dai et al., 2012; Akbari et al., 2013). It is worth mentioning that the formation of anthocyanin-MTE complexes is pH-dependent since it affects the protonation/deprotonation of anthocyanins and consequently their reactivity towards heavy metals.

As for chlorophyll concentrations, the NBI indices (Figure 3) were the lowest for C1, C2 and C4 and statistically different from that of R. In addition, no statistical difference was highlighted between the reference and the cultivars C3 and C5. Defined as an indicator of C:N allocation changes (Cervic

et al., 2012), the NBI could be a simple valuable tool for preventing the rate of senescence of chicory leaves in addition to the chlorophyll measure, and so the premature leaf fall of chicories produced in contaminated soils. This highlight has been suggested in several studies about miscanthus (Mos et al., 2013; Rusinowski et al., 2019).

Despite the short time-experimentation (211 days) and the limited-size of the containers, the exported metal from soil ranged from 0.83 mg to 1.59 mg Pb, 1.44 to 2.03 mg Cd and 6.94 to 10.87 mg Zn. Based on the dataset, a remediation factor (RF) was calculated according to the definition described in Sabir et al. (2015). Briefly, this factor was defined as the ratio of the mass of metal in the set of plants cultivated in each container to soil as follows (Eq. 4):

$$RF = \frac{MTE\ concentration\ in\ plant \times plant\ dry\ biomass}{MTE\ concentration\ in\ soil \times mass\ of\ air-dried\ contaminated\ soil} \times 100 \text{ (eq 4)}$$

It was clearly demonstrated that the mechanism uptake of MTE depended on several parameters like the root zone, the environmental conditions, the soil parameters, the plant species, the speciation of MTE (Tangahu et al., 2011) and so, the bioavailability of MTE. Thus, two remediation factor indexes, named RF_{LMWOAs} and RF_{CaCl_2} were calculated considering the extractable MTE concentrations measured in the mixture of organic acids (LMWOAs) and in the 0.01 M calcium chloride solution. The results are summarized in Table 3. Although it was demonstrated that Pb, Cd and Zn were poorly phytoavailable using miscanthus as plant model (Nsanganwimana et al., 2015; 2021), the RF-Cd values were in the range 0.67-0.95 %, depending on the cultivars. Even if the maturity of chicories has not been reached, this result is interesting since the RF values tend to increase with time (Saraswat and Rai, 2011). According to our knowledge, no study reported similar data using chicory cultivars. In contrast, some RF-Cd values were reported in literature using various plant models in anthropogenically or artificially contaminated soils (Hu et al., 2014; Chhajro et al., 2018; Moemeri and Khalaki, 2019). Chhairo et al. (2018) revealed RF-Cd values of 0.39 without any amendment and up to 1 using chelators (EDTA, NTA, citrate) and castor bean (*Ricinus communis*) whereas Rf-Cd values below 0.7 was highlighted whatever the treatments (EDTA, EGTA, citrate; Sun et al., 2011; Hu et al., 2014). However, RF-Cd values between 1.0 and 4.0 using *Secale montanum* and *Leucaena leucocephala* were reported under a phytoextraction context (Saraswast and Rai, 2009, 2011; Moameri

and Khalaki, 2019) As shown in Table 3, the RF-Pb values are very low, from 0.006 to 0.01. They are in the same order of magnitude as those reported in Moameri and Khalaki, (2019) and Sun et al. (2011). Nevertheless, depending on the ability of shrub and tree species to phytoextract Pb, some authors highlighted medium and high RF-Pb values (from 0.5 to 13.5; Aman et al., 2018; Singh et al., 2020). The RF-Zn values were ten-fold higher than those of RF-Pb although the total concentrations of these MTE in the soil studied were quite similar. Even if similar RF-Zn values were reported in Sun et al. (2001) as those obtained in the current study, it is worth noting that RF-Zn values are between 0.3 to 2.1, depending on plant species and soil characteristics (Aman et al., 2018; Sarawat and Rai, 2011; Sabir et al., 2015).

In the Gentle soil Remediation Options (GRO) the common goal is to decrease the labile pool of MTE because it represents the most available pool of MTE. This pool is a part of the total metal concentration and is commonly named bioavailable. In this concept, phytoextraction and phytoremediation are both strategies that can be used to achieve this objective if this plant-based soil-management is integrated in the bioeconomy concept. From these global considerations, we defined the RF_{LMWOAs} and RF_{CaCl2} described above. Values of these two remediation factors were higher than those of RF since there were defined from the extractable-MTE concentrations. As indicated in Table 3, the $RF_{LMWOAs-Cd}$ is higher than 4 meaning that chicories can be used as phytoextractor for bioavailable-Cd in long run. This result correlated with the conclusion described previously from the BAC, BCF and TF values. In contrast, the results remain less evident for Pb and Zn since their respective RF_{LMWOAs} values were generally below 1. Anyway, chicories are able to decrease the pool of the bioavailable-MTE in short time as shown by the RF_{CaCl2} values for the three MTE studied.

Although the growth period was limited in time due to the size of containers, this new approach and the whole of results obtained from container-experiments should provide new insights into the potential of chicories in lowering the environmental risks after a large-scale chicory production on contaminated agricultural soils. However, it is worth mentioning that further studies need to be conducted due to (i) the restriction of the root size in the current study and not for field studies and (ii) a potential lateral and vertical gradient of metal(loid)s contamination levels in real contaminated sites.

4. Conclusion

This study, carried out over a period of 211 days, was conducted to determine the concentration and time distribution of Cd, Pb and Zn in five cultivars of chicory which have been produced on an agricultural contaminated soil. Leaves and roots analysis at 4 time points during experimentation (at 74, 105, 156 and 211 days, respectively) revealed that the Cd and Zn concentration in plants organs at the end of the experiments were higher in shoots than in roots (up to 7-fold and 3.4-fold higher, respectively) whatever the cultivars were, while no significant differences were observed for Pb. Indeed, Cd-BAC and Cd-TF increase during the growing period to reach values above 4 and 7, highlighting the ability of chicory to reduce the Cd content of the soil. This reasoning is also valid for Zn, but to a lesser extent since the Zn-TF reaches values higher than 3 at 211 days. The Zn-BAC values are lower than 1 from the first harvest to the last one. In addition, MTE concentrations in shoots have been related to the content of chlorophyll, anthocyanins and nitrogen balance index. It appears that an increase of the MTE concentration in the leave induces i) an increase of the anthocyanin content, which may provide to the chicory a better adaptation to contamination level ii) a decrease of the content of chlorophyll and nitrogen balance index, which could be related to the senescence phenomenon. Finally, the phytoextracting power of chicory has been discussed regarding the bioavailable part of the total MTE concentration and has been identified as a good phytoextractor, particularly for Cd. These news results thus highlight the interest of harvesting the aerial parts of chicory thus limiting the deposition of MTE after large-scale chicory production on contaminated agricultural soils.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The authors thank the Region Hauts-de-France (FEDER funding: Fonds européen de développement regional), JUNIA and the Florimond Desprez company for financial support of this work and technical help and facilities. The authors also acknowledge B. Desprez, N. Henry and Y. Delourme for their technical advice.

References

- Ahmed, N.U., Park, J.I., Jung, H.J., Yang, T.J., Hur, Y., Nou, I.S., 2014. Characterization of dihydroflavonol 4-reductase (DFR) genes and their association with cold and freezing stress in *Brassica rapa*. *Gene* 550, 46–55. <https://doi.org/10.1016/j.gene.2014.08.013>
- Akbari, R., Hatamzadeh, A., Sariri, R., Bakhshi, D., 2013. Relationship of flower color parameters and metal ions of petal tissue in fully opened flowers of *Gerbera*. *J. Plant Stud.* 2, 89–96. <https://dx.doi.org/10.5539/jps.v2n1p89>
- Aksoy, A., 2008. Chicory (*Cichorium Intybus* L.): A possible biomonitor of metal pollution. *Pak. J. Bot.* 40, 791–797. <https://doi.org/10.1.1.681.4091>
- Alagić, S.Č., Tošić, S.B., Dimitrijević, M.D., Antonijević, M.M., Nujkić, M.M., 2015. Assessment of the quality of polluted areas based on the content of heavy metals in different organs of the grapevine (*Vitis vinifera*) cv Tamjanika. *Environ. Sci. Pollut. Res.* 22, 7155–7175. <https://doi.org/10.1007/s11356-014-3933-1>
- Ali, H., Khan, E., Sajad, M.A., 2013. Phytoremediation of heavy metals—Concepts and applications. *Chemosphere* 91, 869–881. <https://doi.org/10.1016/j.chemosphere.2013.01.075>
- Alloway, B.J., 2013. Sources of Heavy Metals and Metalloids in Soils, in: Alloway, B.J. (Ed.), *Heavy Metals in Soils: Trace Metals and Metalloids in Soils and Their Bioavailability*, Environmental Pollution. Springer Netherlands, Dordrecht, pp. 11–50. https://doi.org/10.1007/978-94-007-4470-7_2
- Alloway, B.J. (Ed.), 1995. *Heavy Metals in Soils*, 2nd ed. Springer Netherlands.
- Aman, M.S., Jafari, M., Reihan, M.K., Motesarezadeh, Babak., 2018. Assessing some shrub species for phytoremediation of soils contaminated with lead and zinc. *Environ. Earth. Sci.* 77, 1–10. <https://doi.org/10.1007/s12665-018-7256-2>

378 Baker A.J., 1981. Accumulators and excluders — strategies in the response of plants to heavy metals.
 379 J. Plant Nutr. 3, 643–654. <https://doi.org/10.1080/01904168109362867>

380 Bhargava, A., Carmona, F.F., Bhargava, M., Srivastava, S., 2012. Approaches for enhanced
 381 phytoextraction of heavy metals. J. Environ. Manage. 105, 103–120.
 382 <https://doi.org/10.1016/j.jenvman.2012.04.002>

383 Cerovic, Z.G., Masdoumier, G., Ghazlen, N.B., Latouche, G., 2012. A new optical leaf-clip meter for
 384 simultaneous non-destructive assessment of leaf chlorophyll and epidermal flavonoids. Physiol. Plant.
 385 146, 251–260. <https://doi.org/10.1111/j.1399-3054.2012.01639.x>

386 Chalker-Scott, L., 1999. Environmental significance of anthocyanins in plant stress responses.
 387 Photochem. Photobiol. 70, 1–9. <https://doi.org/10.1111/j.1751-1097.1999.tb01944.x>

388 Chhajro, M.A., Rizwan, M.S., Huang, G., Zhu, J., Kubar, K.A., Hu, Hongqing., 2016. Enhanced
 389 accumulation of Cd in castor (*Ricinus communis* L.) by soil-applied chelators. Intern. J. Phytoremed.
 390 18, 664–670. <https://doi.org/10.1080/15226514.2015.1115965>

391 Dai, L.-P., Dong, X.-J., Ma, H.-H., 2012. Antioxidative and chelating properties of anthocyanins in
 392 *Azolla imbricata* induced by cadmium. Pol. J. Environ. Stud. 21, 837–844.

393 Deyris, P.-A., Adler, P., Petit, E., Legrand, Y.-M., Grison, C., 2019. Woody species: a new bio-based
 394 material for dual Ca/Mg catalysis with remarkable Lewis acidity properties. Green Chem. 21, 3133–
 395 3142. <https://doi.org/10.1039/C9GC00770A>

396 Douay, F., Pelfrène, A., Planque, J., Fourrier, H., Richard, A., Roussel, H., Girondelot, B., 2013.
 397 Assessment of potential health risk for inhabitants living near a former lead smelter. Part 1: metal
 398 concentrations in soils, agricultural crops, and homegrown vegetables. Environ. Monit. Assess. 185,
 399 3665–3680. <https://doi.org/10.1007/s10661-012-2818-3>

400 Finnegan, P., Chen, W., 2012. Arsenic Toxicity: The Effects on Plant Metabolism. Front Physiol. 3,
 401 182–190. <https://doi.org/10.3389/fphys.2012.00182>

402 Grant, C.A., Clarke J.M., Duguid, S., Chaney, R.L., 2008. Selection and breeding of plant cultivars to
 403 minimize cadmium accumulation. Sci. Total Environ. 390, 301–310.
 404 <https://doi.org/10.1016/j.scitotenv.2007.10.038>

405 Gomes, H.I., 2012. Phytoremediation for bioenergy: challenges and opportunities. *Environ. Technol.*
 406 *Rev.* 1, 59–66. <https://doi.org/10.1080/09593330.2012.696715>

407 Gong, Y., Zhao, D., Wang, Q., 2018. An overview of field-scale studies on remediation of soil
 408 contaminated with heavy metals and metalloids: Technical progress over the last decade. *Water Res.*
 409 147, 440–460. <https://doi.org/10.1016/j.watres.2018.10.024>

410 Guérin, T., Ghinet, A., Waterlot, C., 2020. Toward a New Way for the Valorization of *Miscanthus*
 411 Biomass Produced on Metal-Contaminated Soils Part 2: *Miscanthus*-Based Biosourced Catalyst:
 412 Design, Preparation, and Catalytic Efficiency in the Synthesis of Moclobemide. *Sustainability* 13, 34.
 413 <https://doi.org/10.3390/su13010034>

414 Hazotte, C., Laubie, B., Rees, F., Morel, J.L., Simonnot, M.-O., 2017. A novel process to recover
 415 cadmium and zinc from the hyperaccumulator plant *Noccaea caerulea*. *Hydrometallurgy* 174, 56–
 416 65. <https://doi.org/10.1016/j.hydromet.2017.09.012>

417 Hechelski, M., Ghinet, A., Louvel, B., Dufrénoy, P., Rigo, B., Daïch, A., Waterlot, C., 2018. From
 418 Conventional Lewis Acids to Heterogeneous Montmorillonite K10: Eco-Friendly Plant-Based
 419 Catalysts Used as Green Lewis Acids. *ChemSusChem* 11, 1249–1277.
 420 <https://doi.org/10.1002/cssc.201702435>

421 Hechelski, M., Louvel, B., Dufrénoy, P., Ghinet, A., Waterlot, C., 2020. Toward a New Way for the
 422 Valorization of *Miscanthus* Biomass Produced on Metal-Contaminated Soils Part 1: Mesocosm and
 423 Field Experiments. *Sustainability* 12, 9370. <https://doi.org/10.3390/su12229370>

424 Hu, Y., Nan, Z., Su, J., Wang, Shengli., 2014. Chelant-Assisted Uptake and Accumulation of Cd by
 425 Poplar from Calcareous Arable Soils Around Baiyin Nonferrous Metal Smelters, Northern China. *Arid*
 426 *Land Res. Manag.* 28, 340–354. <https://doi.org/10.1080/15324982.2013.841305>

427 Janus, A., Pelfrène, A., Heymans, S., Deboffe, C., Douay, F., Waterlot, C., 2015. Elaboration,
 428 characteristics and advantages of biochars for the management of contaminated soils with a specific
 429 overview on *Miscanthus* biochars. *J. Environ. Manage.* 162, 275–289.
 430 <https://doi.org/10.1016/j.jenvman.2015.07.056>

431 Liu, L., Li, W., Song, W., Guo, M., 2018. Remediation techniques for heavy metal-contaminated soils:
 432 Principles and applicability. *Sci. Total Environ.* 633, 206–219.
 433 <https://doi.org/10.1016/j.scitotenv.2018.03.161>
 434 Mandzhieva, S.S., Minkina, T.M., Chaplygin, V.A., Motuzova, G.V., Sushkova, S.N., Bauer, T.V.,
 435 Nevidomskaya, D.G., 2016. Plant contamination by heavy metals in the impact zone of
 436 Novocherkassk Power Station in the south of Russia. *J. Soil. Sediment.* 16, 1383–1391.
 437 <https://doi.org/10.1007/s11368-015-1098-2>
 438 Mirzaei, M., Verrelst, J., Bakhtiari, A.R., Marofi, S., 2021. Potential use of grapevine cv Askari for
 439 heavy metal phytoremediation purposes at greenhouse scale. *Environ. Sci. Pollut. Res.* 28, 12447–
 440 12458. <https://doi.org/10.1007/s11356-020-11129-9>
 441 Moameri, M., Abbasi Khalaki, Masoomah., 2019. Capability of *Secale montanum* trusted for
 442 phytoremediation of lead and cadmium in soils amended with nano-silica and municipal solid waste
 443 compost. *Environ. Sci. Pollut. Res.* 26, 24315–24322. <https://doi.org/10.1007/s11356-017-0544-7>
 444 Mos, M., Banks, S.W., Nowakowski, D.J., Robson, P.R.H., Bridgwater, A.V., Donnison, I.S., 2013.
 445 Impact of *Miscanthus x giganteus* senescence times on fast pyrolysis bio-oil quality. *Bioresour.*
 446 *Technol.* 129, 335–342. <https://doi.org/10.1016/j.biortech.2012.11.069>
 447 Nsanganwimana, F., Pourrut, B., Waterlot, C., Louvel, B., Bidar, G., Labidi, S., Fontaine, J.,
 448 Muchembled, J., Lounès-Hadj Sahraoui, A., Fourier, H., Douay, F., 2015. Metal accumulation and
 449 shoot yield of *Miscanthus x giganteus* growing in contaminated agricultural soils: Insights into
 450 agronomic practices. *Agric. Ecosyst. Environ.* 213, 61–71. <https://doi.org/10.1016/j.agee.2015.07.023>
 451 Nsanganwimana, F., Al Souki, K.S., Waterlot, C., Douay, F., Pelfrène, A., Rodöšková, A., Louvel, B.,
 452 Pourrut, B., 2021. Potentials of *Miscanthus x giganteus* for phytostabilization of trace element-
 453 contaminated soils: *Ex situ* experiment. *Ecotoxicol. Environ. Safe.* 214, 112125.
 454 <https://doi.org/10.1016/j.ecoenv.2021.112125>
 455 Oustriere, N., Marchand, L., Lizama-Allende, K., Roulet, E., Rousset, C., Bordas, F., Mench, M.,
 456 2019. Selection of macrophytes with Cu-enriched root biomass intended for ecocatalyst production.
 457 *Ecol. Eng.* 138, 88–96. <https://doi.org/10.1016/j.ecoleng.2019.07.001>

458 Pelfrêne, A., Kleckeroá, A., Pourrut, B., Nsanganwimana, F., Douay, F., Waterlot, C., 2015. Effect of
 459 miscanthus cultivation bioaccessibility in metal-contaminated soils: comparison between greenhouse
 460 and field experiments. *Environ. Sci. Pollut. Res.* 22, 3043–3054. [https://doi.org/10.1007/s11356-014-](https://doi.org/10.1007/s11356-014-3585-1)
 461 3585-1
 462 Pruvot, C., Douay, F., Hervé, F., Waterlot, C., 2006. Heavy Metals in Soil, Crops and Grass as a
 463 Source of Human Exposure in the Former Mining Areas. *J. Soil. Sediment.* 6, 215–220.
 464 <https://doi.org/10.1065/jss2006.10.186>
 465 Rossini-Oliva, S., Abreu, M.M., Leidi, E.O., 2018. A review of hazardous elements tolerance in a
 466 metallophyte model species: *Erica andevalensis*. *Geoderma* 319, 43–51.
 467 <https://doi.org/10.1016/j.geoderma.2017.12.035>
 468 Rusinowski, S., Krzyzak, J., Clifton-Brown, J., Jensen, E., Mos, M., Webster, R., Sitko, K., Pogrzeba,
 469 M., 2019. New Miscanthus hybrids cultivated at a polish metal-contaminated site demonstrate high
 470 stomatal regulation and reduced shoot Pb and Cd concentrations. *Environ. Pollut.* 252, 1377–1387.
 471 <http://creativecommons.org/licenses/by-nc-nd/4.0/>
 472 Sabir, M., Ali, A., Zia-Ur-rehman, M., Hakeem, K.R., 2015. Contrasting Effects of Farmyard Manure
 473 (FYM) and Compost for Remediation of Metal Contaminated Soil. *Int. J. Phytorem.* 17, 613–21.
 474 <https://doi.org/10.1080/15226514.2014.898019>
 475 Saraswat, S., Rai, J.P.N., 2011. Prospective Application of *Leucaena Leucocephala* for
 476 Phytoextraction of Cd and Zn and Nitrogen Fixation in Metal Polluted Soils. *Int. J. Phytorem.* 13,
 477 271–288. <https://doi.org/10.1080/15226514.2010.483259>
 478 Saraswat, S., Rai, J.P.N., 2009. Phytoextraction potential of six plant species grown in multimetal
 479 contaminated soil. *Chem; Ecol.* 25, 1–11. <https://doi.org/10.1080/02757540802657185>
 480 Savio, M., Cerutti, S., Martinez, L.D., Smichowski, P., Gil, R.A., 2010. Study of matrix effects and
 481 spectral interferences in the determination of lead in sediments, sludges and soils by SR-ETAAS using
 482 slurry sampling. *Talanta* 82, 523–527. <https://doi.org/10.1016/j.talanta.2010.05.017>
 483 Sêkara, A., Poniedzia, M., 2005. Cadmium and Lead Accumulation and Distribution in the Organs of
 484 Nine Crops: Implications for Phytoremediation. *Pol. J. Environ. Stud.* 14, 509–516.

485 Sheoran, V., Sheoran, A.S., Poonia, P., 2016. Factors Affecting Phytoextraction: A Review.
 486 Pedosphere 26, 148–166. [https://doi.org/10.1016/S1002-0160\(15\)60032-7](https://doi.org/10.1016/S1002-0160(15)60032-7)
 487 Shi, G.R., Su, G., Lu, Z., Liu, C.F., Wang, X., 2014 Relationship between biomass, seed components
 488 and seed cd concentration in various peanut (*Arachis hypogaea* L.) cultivars grown on Cd-
 489 contaminated soils. Ecotoxicol. Environ. Saf. 110, 174–181.
 490 <https://doi.org/10.1016/j.ecoenv.2014.09.003>
 491 Shtangeeva, I., Bali, R., Harris, A., 2011. Bioavailability and toxicity of antimony. J. Geochem.
 492 Explor. 110, 40–45. <https://doi.org/10.1016/j.gexplo.2010.07.003>
 493 Sterckeman, T., Douay, F., Proix, N., Fourrier, H., Perdrix, E., 2002. Assessment of the contamination
 494 of cultivated soils by eighteen trace elements around smelters in the north of France. Water Air Soil
 495 Pollut. 135, 173-194. <https://doi.org/10.1023/A:1014758811194>
 496 Singh, B., Kaur, B., Singh, D., 2020. Assessment of different multipurpose tree species for
 497 phytoextraction of lead from lead-contaminated soils. Biorem. J. 24, 215–
 498 230. <https://doi.org/10.1080/10889868.2020.1811634>
 499 Sun, Y.B., Sun, G.H., Zhou, Q.X., Xu, Y.M., Wang, L., Liang, X.F., Sun, Y., Qing, X., 2011.
 500 Induced-phytoextraction of heavy metals from contaminated soil irrigated by industrial wastewater
 501 with Marvel of Peru (*Mirabilis jalapa* L.). Plant Soil Environ. 57, 364–
 502 371. <https://doi.org/10.17221/148/2011-PSE>
 503 Tangahu, B.J., Abdullah, S.R.S., Basri, H., Idris, M., Anuar, N., Mukhlisin, M., 2011. A review on
 504 heavy metals (As, Pb, and Hg) uptake by plants through phytoremediation. International Journal of
 505 Chem. Eng. 1–31. <https://doi.org/10.1155/2011/939161>
 506 Uzu, G., Sobanska, S., Sarret, G., Muñoz, M., & Dumat, C. 2010. Foliar Lead uptake by lettuce
 507 exposed to atmospheric fallouts. Environ. Sci. Technol., 44, 1036–1042.
 508 <https://doi.org/10.1021/es902190u>
 509 Vamerali, T., Bandiera, M., Mosca, G., 2010. Field crops for phytoremediation of metal-contaminated
 510 land. A review. Environ. Chem. Lett. 8, 1–17. <https://doi.org/10.1007/s10311-009-0268-0>

511 Waterlot, C., Bidar, G., Pelfrène, A., Roussel, H., Fourrier, H., Douay, F., 2023. Contamination,
 512 fractionation and availability of metals in urban soils in the vicinity of former lead and zinc smelters,
 513 France. *Pedosphere* 23, 143–159. [https://doi.org/10.1016/S1002-0160\(13\)60002-8](https://doi.org/10.1016/S1002-0160(13)60002-8)
 514 Waterlot, C., Douay, F., 2009. The problem of arsenic interference in the analysis of Cd to evaluate its
 515 extractability in soils contaminated by arsenic. *Talanta* 80, 716–722.
 516 <https://doi.org/10.1016/j.talanta.2009.07.053>
 517 Waterlot, C., Dufrénoy, P., Hechelski, M., Louvel, B., Daïch, A., Ghinet, A., 2019. Benefits of
 518 Ryegrass on Multicontaminated Soils Part 2: A Green Process to Provide Idroclamide. *Sustainability*
 519 11, 6685. <https://doi.org/10.3390/su11236685>
 520 Waterlot, C., Hechelski, M., 2019. Benefits of Ryegrass on Multicontaminated Soils Part 1: Effects of
 521 fertilizers on bioavailability and accumulation of metals. *Sustainability* 11, 5093.
 522 <https://doi.org/10.3390/su11185093>
 523 Waterlot, C., Pruvot, C., Marot, F., Douay, F., 2017. Impact of a phosphate amendment on the
 524 environmental availability and phytoavailability of Cd ad Pb in moderetaly and highly carbonated
 525 kitchen garden soils. *Pedosphere* 27, 588–605. [https://doi.org/10.1016/S1002-0160\(17\)60354-0](https://doi.org/10.1016/S1002-0160(17)60354-0)
 526 Wang, Z., Liu, X., Qin, H., 2019. Bioconcentration and translocation of heavy metals in the soil-plants
 527 system in Machangqing copper mine, Yunnan Province, China. *J. Geochem. Explor.* 200, 159–166.
 528 <https://doi.org/10.1016/j.gexplo.2019.02.005>
 529 Wojciechowska, N., Sobieszczuk-Nowicka, E., Bagniewska-Zadworna, A., 2018. Plant Organ
 530 senescence – regulation by manifold pathways. *Plant Biol.* 20, 167–181. <http://doi:10.1111/plb.12672>
 531 Wolf, M., Paulino, A.T., 2020. Phytotoxicity increase induces by zinc accumulation in *Cichorium*
 532 *intybus*. *Bull. Environ. Contam. Toxicol.* 105, 405–410. <https://doi.org/10.1007/s00128-020-02960-4>
 533 Zeng, H., Chen, L., Zhou, X., Zeng, Q., 2019. Cadmium accumulation in winter crops and the
 534 assessment of paddy soil phytoremediation in southern China. *Environ. Sci. Pollut. Res.* 26, 17173–
 535 17182. <https://doi.org/10.1007/s11356-019-05054-9>
 536 Zehra, A., Sahito, Z.A., Tong, W., Tang, L., Hamid, Y., Wang, Q., Cao, X., Khan, M.B., Hussain, B.,
 537 Jatoi, S.A., He, Z., 2020. Identification of high cadmium-accumulating oilseed sunflower (*Helianthus*

538 *annuus*) cultivars for phytoremediation of an Oxisol and an Inceptisol. *Ecotoxicol. Environ. Saf.* 187,
539 109857. <http://doi:10.1016/j.ecoenv.2019.109857>

540 Zhou, Z., Guo, Y., Hu, L., He, L., Xu, B., Huang, Z., Wang, G., Chen, Y., 2020. Potential use of king
541 grass (*Pennisetum purpureum* Schumach. × *Pennisetum glaucum* (L.) R.Br.) for phytoextraction of
542 cadmium from fields. *Environ. Sci. Pollut. Res.* 27, 35249–35260. [https://doi.org/10.1007/s11356-](https://doi.org/10.1007/s11356-020-09844-4)
543 020-09844-4

544

Table 1 Mean values of bioaccumulation factor (BAC), bioconcentration factor (BCF) and translocation factor (TF)

Cultivar	Time (days)	BAC			BCF			TF		
		Pb	Cd	Zn	Pb	Cd	Zn	Pb	Cd	Zn
C1	74	0.01	2.06	0.15	0.13	1.05	0.16	0.11	1.97	0.95
	105	0.02	2.14	0.13	0.02	0.44	0.05	0.81	4.91	2.54
	156	0.01	2.45	0.17	0.01	0.39	0.03	1.44	6.31	5.37
	211	0.01	3.29	0.21	0.01	0.57	0.05	0.89	5.80	4.47
C2	74	0.02	1.93	0.15	0.17	1.09	0.20	0.10	1.77	0.75
	105	0.03	1.77	0.11	0.07	0.44	0.04	0.42	4.06	2.62
	156	0.02	2.89	0.15	0.01	0.33	0.04	3.36	8.70	4.22
	211	0.01	3.22	0.20	0.01	0.62	0.05	1.04	5.17	3.63
C3	74	-	-	-	-	-	-	-	-	-
	105	0.02	2.03	0.12	0.05	0.60	0.09	0.45	3.38	1.40
	156	0.02	2.28	0.14	0.01	0.31	0.03	2.48	7.45	4.10
	211	0.02	2.55	0.16	0.01	0.56	0.05	1.34	4.57	3.33
C4	74	0.04	1.90	0.14	0.22	1.15	0.18	0.16	1.65	0.77
	105	0.02	2.23	0.11	0.03	0.57	0.07	0.66	3.90	1.73
	156	0.01	2.73	0.15	0.00	0.09	0.05	3.53	28.82	2.90
	211	0.01	4.69	0.20	0.01	0.70	0.06	0.89	6.65	3.38
C5	74	0.01	2.06	0.14	0.13	1.14	0.18	0.10	1.81	0.78
	105	0.01	1.57	0.11	0.03	0.50	0.06	0.44	3.12	1.90
	156	0.02	2.26	0.13	0.01	0.30	0.03	2.02	7.49	4.05
	211	0.01	3.58	0.15	0.01	0.49	0.05	0.86	7.38	2.97

Table 2 Leaf accumulation and root uptake of MTE by chicories

cultivar	MTE accumulation in leaves			MTE uptake by roots			MTE uptake by chicories		
	Cd (mg)	Pb (mg)	Zn (mg)	Cd (mg)	Pb (mg)	Zn (mg)	Cd (mg)	Pb (mg)	Zn (mg)
C1	1.18	0.28	6.23	0.41	0.56	2.72	1.59	0.83	8.96
C2	1.42	0.33	6.65	0.51	0.51	4.21	1.93	0.84	10.87
C3	1.02	0.54	4.94	0.57	1.05	3.91	1.58	1.59	8.86
C4	1.44	0.39	5.04	0.59	0.67	4.51	2.03	1.06	9.56
C5	1.01	0.30	3.40	0.43	0.93	3.54	1.44	1.23	6.94

Table 3 Remediation factors (RF)

cultivar	RF			RF _{LMWOAs}			RF _{CaCl2}		
	Cd	Pb	Zn	Cd	Pb	Zn	Cd	Pb	Zn
C1	0.74	0.006	0.05	4.42	0.79	0.26	28.4	119.2	80.5
C2	0.90	0.006	0.06	5.37	0.79	0.32	34.5	119.6	97.7
C3	0.74	0.010	0.05	4.40	1.50	0.26	28.3	227.0	79.6
C4	0.95	0.007	0.06	5.65	1.00	0.28	36.3	152.0	85.9
C5	0.67	0.008	0.04	4.00	1.16	0.20	25.7	175.8	62.4

Fig. 1. Pb, Cd and Zn accumulation (A,B,C) in the roots and shoots depending on the growing time and cultivars

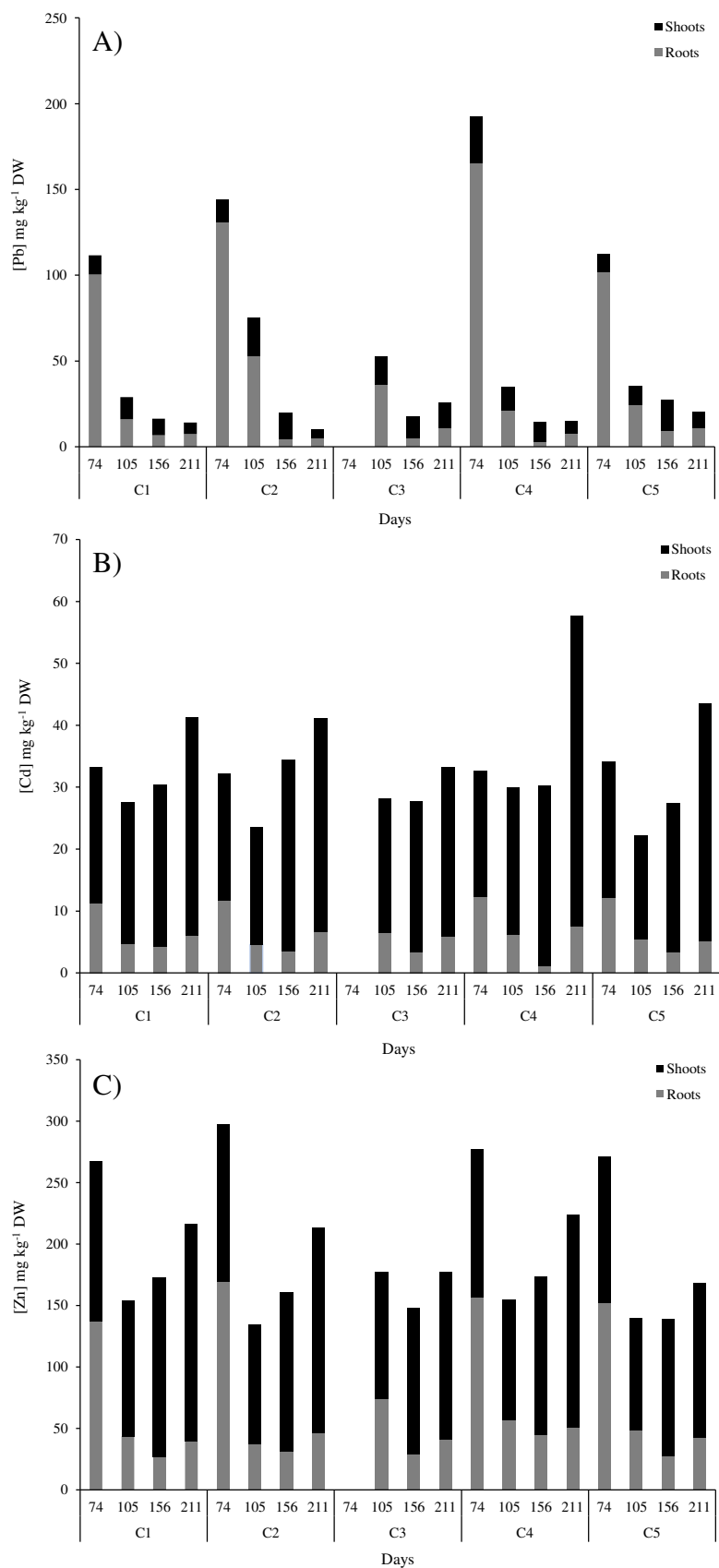


Fig. 2. Distribution (%) of leaf and root biomass for the five cultivars studied

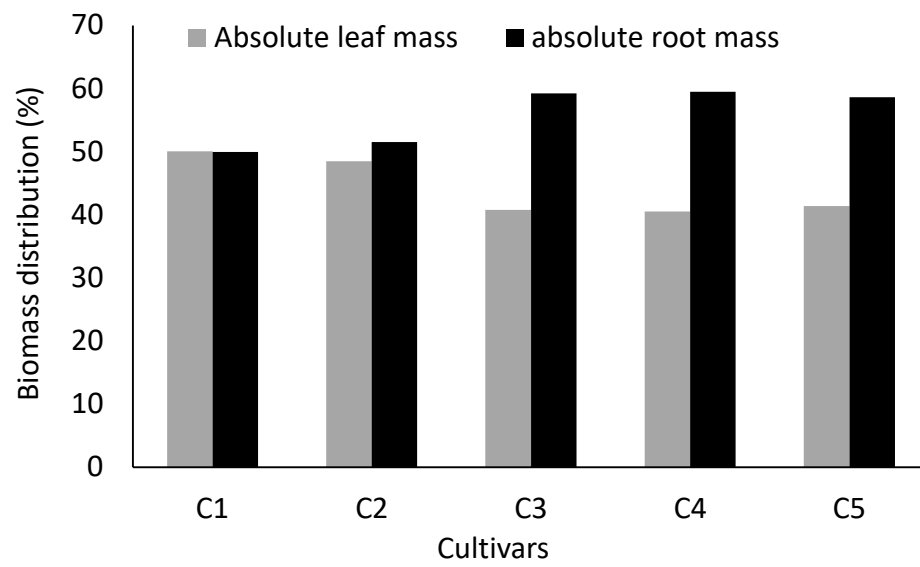
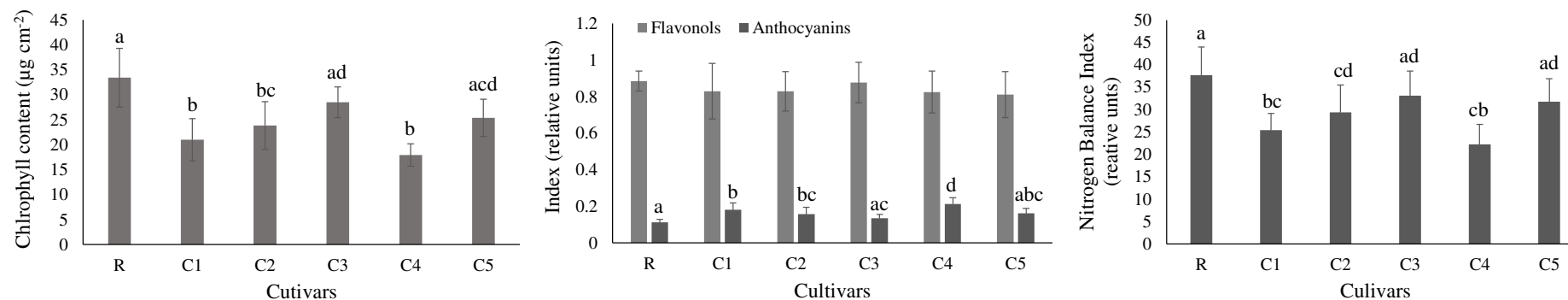


Fig. 3. Chlorophyll content, Flavonols index, Anthocyanins index and Nitrogen Balance Index measured in leaves of chicories (reference and cultivars)



Letters (a, b, c, d, e) refer to significant differences ($p < 0.05$) between reference and the five cultivars studied. Values are means \pm standard deviation ($n=20$).

Root chicory as selective MTE phytoextractor and high root-leaf translocation of Cd

