DE GRUYTER OPEN

Alena Vollmannova*, Eva Margitanova*, Tomas Toth*, Radovan Stanovic*, Tatiana Bojnanska**, Iveta Cicova***, Michaela Benkova***

Differences in ability of selected amaranth cultivars to accumulate risky metals¹

Zróżnicowana zdolność wybranych odmian szarłatu (Amaranhtus) do pobierania i akumulacji metali ciężkich z gleb

* Prof. RNDr. Alena Vollmannova, PhD.; Ing. Eva Margitanova, PhD.; Assoc. Prof. RNDr. Ing. Tomas Toth, PhD.; Ing. Radovan Stanovic, PhD., Dep. of Chemistry, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, 949 76 Nitra, Slovak Republic; tel.: +421 37 641 4374; e-mail: alena.vollmannova@uniag.sk

** Assoc. Prof. Ing. Tatiana Bojnanska, CSc., Dept. of Storing and Processing of Plant Products, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, 949 76 Nitra, Slovak Republic

Keywords: amaranth, heavy metals, aboveground biomass, phytoremediation Słowa kluczowe: szarłat, metale ciężkie, nadziemna biomasa, fitoremediacja

Abstract

The objective of the study is to compare the degree of accumulation of risky metals by seeds and aboveground biomass of five amaranth cultivars cultivars (Golden Giant, Rawa, Annapurna, Oscar Blanco, Koniz), as well as to assess the metal input extent in system soil-plant. The soil of amaranth cultivation was uncontaminated with the exception of Cd and Pb contents, those were 40% and 10% higher than limits given for the soil extract by aqua regia and by NH₄NO₃, respectively. In seeds of all investigated amaranth cultivars the maximal allowed amounts for Cd and Pb were by 60-100% and 25-200% (respectively) exceeded. In aboveground amaranth biomass 0.17-12.25 fold higher amounts of heavy metals were determined in comparison to amaranth seeds. Our results confirm, that amaranth seeds and leaves as food raw materials could represent a risk to the health of the consumer from the aspect of high Cd and Pb amounts. Because of the ability to produce abundance of biomass and at the same time to accumulate high amounts of dangerous heavy metals amaranth could be used as a potential plant for a soil phytoremediation. The results confirmed the ability of amaranth to accumulate metals even from relatively "clean" soil.

© IOŚ-PIB

1. INTRODUCTION

Amaranth is an 8000-year-old cultivated crop originating in the American continent. It is not demanding on soil and climatic conditions, is resistant to stress and high temperatures and produces a lot of biomass. Amaranth is a very versatile crop that is grown in a wide range of agroclimatic conditions; it resists drought, heat, and pests, and adapts readily to new environments [Rana et al. 2007]. Cultivated amaranth species can be used, not only as a source of edible seeds, leafy vegetables, and forage, but also as ornamentals [Mlakar et al. 2009]. In West Africa

Streszczenie

Celem przeprowadzonych badań było porównanie stopnia akumulacji metali ciężkich (mogących stanowić ryzyko dla zdrowia ludzi) przez nasiona i biomasę pięciu wybranych odmian uprawnych szarłatu (Golden Giant, Rawa, Annapurna, Oscar Blanco, Koniz). Celem pracy była także ocena zależności pobierania metali ciężkich: gleba-roślina. Stwierdzono, że badane gleby były zanieczyszczone Cd i Pb, których zawartość przekraczała odpowiednio 40% i 10% zawartości wyjściowej oznaczonej w ekstrakcie wody królewskiej i NH₄NO. W nasionach Szarłatu wszystkich badanych odmian zawartość Cd i Pb przekraczały dozwolone zawartości tych pierwiastków odpowiednio o 60-100% i 25-200%. W częściach nadziemnych szarłatu zawartość badanych metali była średnio 0,17-12,25 krotnie wyższa niż w nasionach badanych roślin. Na podstawie otrzymanych wyników stwierdzono, że nasiona oraz liście szarłatu wykorzystywane na cele żywnościowe mogą stanowić zagrożenie dla zdrowia konsumentów, wynikające z wysokich zawartości Cd i Pb. Szarłat posiadając dużą zdolność akumulacji Cd i Pb, poprzez wytwarzanie obfitej biomasy roślinnej może być wykorzystany w celu fitoremediacji gleb. Wyniki potwierdziły zdolność do akumulacji metali ciężkich przez różne odmiany szarłatu nawet z terenów uznawanych za niezanieczyszczone.

amaranth is cultivated also for its edible leaves rich in vitamins and dietary minerals. On the other hand, amaranth plant takes from the soil both essential and toxic elements over a wide range of concentrations [Türkdoğan et al. 2003]. Phytoremediation is a cost-effective technology for environmental cleaning if native plants were applied in each polluted areas [Chehregani et al. 2009]. There are numerous mechanisms by which plants may remediate contaminated sites such as phytoaccumulation, phytoextraction, phytostabilisation, phytotransformation,

¹This work was co-funded by European Community under project No. 26220220180: Building Research Centre "AgroBioTech" and also supported by project: VEGA 1/0308/14.

*** Ing. Iveta Cicova, PhD., Ing. Michaela Benkova, PhD., National Agriculture and Food Centre, Plant Production Research Centre in Piestany, Slovak Republic phytovolatilisation and rhizodegradation [Susarla et al. 2002]. Phytoextraction may provide an attractive alternative for the clean up of heavy metal-contaminated soils [Shivhare, Sharma 2012]. According to Ziarati and Alaedini [2014] amaranth (*Amaranthus* sp.) may probably be employed as a potent phytoremediation of heavy metals from polluted soils and can be cultivated or grown easily in contaminated soils.

The objective of the study is to compare the degree of accumulation of risk metals by amaranth seeds and aboveground biomass and to evaluate the safety of consumption parts of five different amaranth cultivars from the aspect of heavy metal content. Soil as a starting place for the input of risk substances into a human food chain was analysed, too.

2. MATERIAL AND METHODS

Soil

In soil the agrochemical characteristics (pH/KCl, content of K, Mg, P, Ca and humus) were determined. Exchange soil reaction pH/ KCl (c(KCl) = 1 mol·dm⁻³, CentralChem, Slovakia) was determined electrometrically (691 pH Meter Metrohm, Swiss), a content of organic carbon (C_{ox} , %) was determined using volumetric method according to Tjurin (H₂SO₄: Merck, Germany, K₂Cr₂O₇: Merck, Germany; (NH₄)₂Fe(SO₄)₂•6H₂O: Merck, Germany) while a content of humus (Hum, %) was calculated from C_{ox} content. Contents of available nutrients (K, Mg, Ca, P) were determined using Mehlich 3 extraction by AAS method (K, Mg, Ca) or spectrophometrically (P).

Pseudototal content of risk metals (Zn, Cu, Ni, Cr, Pb and Cd) including all the form besides residual metal fraction was assessed in soil extract by *aqua regia* (HCI: CentralChem, Slovakia, HNO₃: Merck, Germany) and content of mobile forms of selected heavy metals in soil extract by NH_4NO_3 (c = 1 mol·dm⁻³, Merck, Germany). Gained results were evaluated according Law No. 220/2004 (valid in the Slovak Republic) as well as threshold values proposed EC (2006). Used analytical method was flame AAS (AAS Varian AA Spectr DUO 240 FS/240Z/UltrAA, Varian, Australia).

Plant material

The aboveground biomass of five amaranth cultivars (Golden Giant, Rawa, Annapurna, Oscar Blanco, Koniz) were manually separated, dried at 105 °C to a constant weight and pulverized (Grindomix 200 GD, Retsch, Germany). Plant samples (1 g) were mineralised in a closed system of microwave digestion (Mars X-Press 5, CEM Corp., USA) in a mixture of 5 cm³ HNO₃ (Suprapur, Merck, Germany) and 5 cm³ deionised (0.054 μ S·cm⁻¹) water (Simplicity 185, Millipore, UK). The digested substances were subsequently filtered through a quantitative filter paper Filtrak 390 (Munktell, Germany) and filled up with deionised water to a volume of 50 cm³. Contents of risky metals (Zn, Cu, Ni, Cr, Pb and Cd) were determined by AAS method (AAS Varian AA Spectr DUO 240FS/240Z/UltrAA, Varian, Australia) and expressed as mg/kg DM (dry matter).

Statistical analysis

Statistica 8 (StatSoft, Inc.) was used for the statistical processing of the results. One-way ANOVA was used. Mean comparisons between cultivars were done by the Fisher's LSD (least significant difference) test.

3. RESULTS AND DISCUSSION

The determined soil reaction is neutral, contents of bioavailable nutrients are low (P), medium (K) and high (Mg). Humus supply is good (Table 1).

The soil of amaranth cultivation was uncontaminated (Table 2). Only determined Cd content was by 40% and Pb content by 10% higher than limit values given by Law No. 220/2004 for the soil extract by aqua regia and for the soil extract by NH_4NO_3 , respectively. But on the other hand the determined Cd content did not exceed the threshold value given by EC (2006).

In aboveground amaranth biomass (Table 3) the followed values of heavy metal content were determined (in mg·kg⁻¹ DM): Zn: 26.20-30.38; Cu: 6.20-16.84; Ni: 0.63-1.75; Cr: 3.90-8.00; Pb: 0.70-2.00; Cd: 0.69-1.87. According to Sola et al. [2003] metal accumulation in the plant tissues of Amaranthus was found to be proportionate to the level of soil concentrations for Pb while Cd level in the crop tissues exceeded that in the soil. Ko et al. [2014] suggested that the plants of Amaranthus could be hyperaccumulators of several metals, and were particularly effective in accumulating Zn and Cu. Pb and Cd, while accumulated in measurable amounts, did not decrease significantly in the soil, suggesting these species may not be the best choice for plants to be used when bioremediating Pb- or Cd-contaminated areas. Abubakar et al. [2014] confirmed a negative relationship between the concentration of Pb in the soil and that in the stem and leaves of amaranth. Li et al. [2012] found that Cd removal from soil was enhanced by the hypoaccumulator Amaranthus hypocondriacus after NPK or NP fertilisation due to an increase on plant biomass. According to Ondo et al. [2012] Amaranthus cruentus plant can accumulate higher concentrations of Cu and Zn if the cultivated soil contains moderately higher Cu and Zn levels.

The statistically significant differences in values of heavy metal content measured in in aboveground biomass between investigated amaranth cultivars were confirmed.

To characterise quantitatively the transfer of an element from soil to plant, the soil–plant Partition Coefficient or Transfer Factor (TF) or Concentration Ratio or Biological Accumulation Coefficient (BAC) that expresses the ratio of contaminant concentration in plant parts to concentration in dry soil is used [Chojnacka et al. 2005]. In Table 4, the calculated values of TF are presented.

The higher the value of the TF, the more mobile/available the metal is. Generally, the transfer factors were low. This may be because only the accumulation of metals in the leaves were studied more metals could have accumulated in the root [Olayinka et al. 2011]. These authors determined in *Amaranthus* leaves the contents of Cd, Cr, Pb and Zn (1.4–5.2; 43.3–96.0; 20.0–49.5 and 78.1–112.0 mg/kg DM, respectively). The calculated values of TF 0.06–0.15 (Cd), 0.09–0.15 (Cr), 0.02–0.08 (Pb) and 0.11 (Zn) were lower in comparison to our results.

Table 1. Agrochemical characteristics of soil

Agrochemical characteristics	pH/H ₂ O	₂O pH/KCI	Сох	Humus	N*	K *	Ca*	Mg*	P*
	ph/h ₂ O		(%)		(mg/kg DM)				
	8.17	7.25	1.39	2.39	3145	295	3803	327	26.8

Table 2. The content of heavy metals in soil (mg·kg⁻¹ DM)

Heavy metals	Cu	Zn	Cd	Pb	Cr	Ni
Aqua regia	25.2	74.2	0.98	26.2	32.6	44.6
Limit value*	60	150	0.7	70	70	50
Threshold value**	100	200	1.5	100	100	70
NH_4NO_3 (c = 1 mol/dm ³)	0.085	0.025	0.026	0.11	0.06	0.16
Critical value*	1	2	0.1	0.1	-	1.5

*Law 220/2004

**European Commission (2006)

Table 3. Content of heavy metals in aboveground amaranth biomass (mg·kg⁻¹ DM)

Amaranth cultivar	Zn	Cu	Ni	Cr	Pb	Cd
Golden Giant	26.63 ^{a,b}	9.87 ^b	0.63 ª	3.90 ª	0.70 ª	0.69 ª
Rawa	28.83 ^{a,b}	8.60 ^b	1.25 ^b	8.00 ^b	1.83 ^b	1.43 ^b
Annapurna	30.38 b	9.40 ^b	1.75 °	7.70 ^b	2.00 b	1.55 ^{b,c}
Oscar Blanco	26.20 ^{a,b}	6.20 ª	1.28 ^b	4.80 ª	1.83 ^b	1.74 ^{c,d}
Koniz	25.70 ª	16.84 °	1.40 ^b	4.90 ª	0.70 ª	1.87 ^d
P-value	0.1500	0.0000	0.0000	0.0000	0.0000	0.0000
F-ratio	1.90	43.27	14.53	29.00	57.47	29.68

Average values marked with the same letter are not significantly different (p< 0.05)

Table 4. Ratio of concentration of metal in plants to metal in soil (Transfer factors)

Amaranth cultivar	Zn	Cu	Ni	Cr	Pb	Cd
Golden Giant	0.359	0.391	0.014	0.120	0.027	0.704
Rawa	0.388	0.341	0.028	0.245	0.070	1.459
Annapurna	0.409	0.373	0.039	0.236	0.076	1.582
Oscar Blanco	0.353	0.246	0.029	0.147	0.070	1.776
Koniz	0.346	0.668	0.031	0.150	0.027	1.908

4. CONCLUSION

Our results confirm, that amaranth leaves as food raw materials could represent a risk to the health of the consumer from the aspect of high Cd and Pb amounts. Because of the ability to produce an abundance of biomass and at the same time to accumulate high amounts of risk heavy metals amaranth could be used as a potential plant for a soil phytoremediation. The results confirmed the ability of amaranth to accumulate risk metals even from relatively "clean" soil.

REFERENCES AND LEGAL ACTS

- ABUBAKAR M.M., ANKA U. S., AHMAD M. M., GETS B.U. 2014. The Potential of *Amaranthus caudatus* as a Phytoremediating Agent for Lead. Journal of Environment and Earth Science, 4: 121–124.
- CHEHREGANIA., NOORIM., YAZDIH.L. 2009. Phytoremediation of heavy-metal-polluted soils: Screening for new accumulator plants in Angouran mine (Iran) and evaluation of removal ability. Ecotoxicology and Environmental Safety, 72: 1349– 1353.
- CHOJNACKA K., CHOJNACKI A., GORECKA H. 2005. Bioavailability of potentially toxic metals from polluted soils to plants. Science of the Total Environment, 337: 175–182.
- EC No. 1881/2006. Commision Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. URL: http://eur-lex.europa.eu/ LexUriServ/LexUriServ.do?uri=OJ:L:2006:364:0005:0024:EN :PDF (accessed 24/02/2015).
- KO C.-H., CHANG F.-C., WANG, Y.-N., CHUNG C.-Y. 2014. Extraction of Heavy Metals from Contaminated Soil by Two Amaranthus spp. Clean Soil Air Water, 42: 635–640.
- Law No. 220/2004 (2004) On the Protection and Use of Agricultural Land. National Council of Slovak Republic, Bratislava. URL: http://www.lecol.sk/images/stories/ lecol/220_2004.pdf (accessed 17/03/2015).
- LI N.Y., FU Q.L., ZHUANG P., GUO B., ZOU B., LI Z.A. 2012. Effect of fertilizers on Cd uptake of Amaranthus Hypochondriacus, a high biomass, fast growing and easily cultivated potential Cd hyperaccumulator. International Journal of Phytoremediation, 14: 162–173.
- MLAKAR S.G., TURINEK M., JAKOP M., BAVEC M., BAVEC F. 2009. Nutrition value and use of grain amaranth: Potential future application in bread making. Agricultura, 6: 43–53.

- OLAYINKA K.O., OYEYIOLA A.O., ODUJEBE F.O., OBOH B. 2011. Uptake of potentially toxic metals by vegetable plants grown on contaminated soil and their potential bioavailability using sequential extraction. Journal of Soil Science and Environmental Management, 2(8): 220–227.
- ONDO J.A., BIYOGO R.M., ABOGO MEBALE A.J., EBA F. 2012. Pot experiment of the uptake of metals by Amaranthus cruentus grown in artificially doped soils by copper and zinc. Food Science and Quality Management, 9: 28–33.
- RANA J.C., PRADHEEP K., YADAV S.K., VERMA V.D., SHARMA P.C. 2007. Durga: A new variety of grain amaranth for cultivation in hill regions. Indian Farming, 57(1): 27–28.
- SHIVHARE L., SHARMA S. 2012. Effect of Toxic Heavy Metal Contaminated Soil on an Ornamental Plant Georgina wild (Dahlia). Journal of Environmental & Analytical Toxicology, 2: 156.
- SOLA O., RASHEED O.A., TALIAT O. 2003. Urban agricultural production: heavy metal contamination of Amaranthus cruentus L. grown on domestic refuse landfill soils in Ibadan, Nigeria. Emirates journal of food and agriculture,15 (2): 87-94.
- SUSARLA S., MEDINA V.F., MC CUTCHEON S.C. 2002. Phytoremediation: an ecological solution to organic chemical contamination. Ecological Engineering, 18: 647–658
- TÜRKDOĞAN M.K., KILICEL F., KARA K., TUNCER I., UYGAN I. 2003. Heavy metals in soil, vegetables and fruit in the endemic upper gastrointestinal cancer region of Turkey. Environmental Toxicology and Pharmacology, 13: 175–179.
- ZIARATI P., ALAEDINI S. 2014. The Phytoremediation Technique for Cleaning up Contaminated Soil By *Amaranthus* sp. Journal of Environmental & Analytical Toxicology, 4: 208.