ECOLOGICAL CHEMISTRY AND ENGINEERING A

Vol. 18, No. 9–10 2011

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HEAVY METAL CONTENTS AND THE SANITARY STATE AS AN ASSESSMENT OF RADISH (*Raphanus sativum* L.) QUALITY

ZAWARTOŚĆ METALI CIĘŻKICH I STAN SANITARNY JAKO OCENA JAKOŚCI RZODKIEWKI (*Raphanus sativum* L.)

Abstract: The research aimed at an assessment of health quality of radish originating from Krakow open air markets. The health quality was estimated on the basis of an analysis of the content and distribution of heavy metals in radish as well as its sanitary state. Heavy metal contents (Zn, Cu, Cd, Pb, Cr and Ni) were assessed in radish flesh, skin and leaves after dry mineralization and the ash dissolving in HNO_3 using ICP-EAS method. Microbiological analyses comprised determining *Coli* and *Salmonella* bacteria count as well as the number of anaerobic and spore forming bacteria (*Clostridium perfringens*).

Heavy metal contents in the studied radish fluctuated widely depending on the analyzed part. The highest contents of zinc, copper, chromium, lead and cadmium were assessed in leaves, lower in the skin and the lowest in radish flesh. Tested radish did not meet the consumer standards for zinc and cadmium. Presence of *Coli* bacteria was noted in 36 % of samples, *Salmonella* bacteria in 10 % and *Clostridium perfringens* only in 2 %.

Keywords: radish, heavy metals, sanitary state

Contamination of foods of plant origin poses a serious health problem. Lead and cadmium are counted among the main food pollutants which create the gravest hazard to human health, both because of their toxicological properties and common occurrence [1]. Zinc, copper, nickel, manganese and iron which in some pre-determined amounts are crucial for the proper course of physiological processes in living organisms, in excessive quantities may constitute a threat to human health [1–3]. Vegetables are valuable element of human diet providing a supplement of mineral salts and vitamins. Therefore, it is necessary to monitor vegetable quality focusing on the presence of

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potentially harmful substances [2, 3]. As has been stated by numerous authors, vegetable plants cultivated in soils and substrata polluted with heavy metals accumulate their excessive quantities which worsens the quality of the plants [4, 5]. Moreover, it has been demonstrated that in the regions under the influence of industrial emission and close to large city agglomerations heavy metal contents in both soil and plants raise considerably.

The research aimed at an assessment of health quality of radish originating from the open air markets in Krakow. Radish quality was assessed on the basis of the content and distribution of heavy metals and radish sanitary state.

Material and methods

The research was conducted at the turn of April and May 2008 on radish (*Raphanus sativus* L.). The studies covered 5 open air markets situated in Krakow. In most cases radish originated from horticultural and agricultural holdings located about 20–30 km from Krakow city agglomeration. 40 samples of radish were selected randomly for the tests. The purchased plant material was washed and divided into leaves, skin and flesh which were then dried at 80 ^{o}C in a drier with forced air flow. After drying the plant material was crushed and dry-mineralized. The ash was dissolved in HNO_3 acid in 1:2 ratio and in the obtained solution zinc, copper, nickel, chromium, lead and cadmium concentrations were assessed using *atomic emission spectrometry with inductively coupled argon plasma* (ISP-AES) on JY 238 ULTRACE apparatus (Jobin Von Emission). Microbiological tests were conducted to determine *Coli* count, *Salmonella* count and anaerobic spore forming bacteria (*Clostridium perfringens*) number [6–9]. For this purpose from each radish bunch stem and root nodosities were cut off from the leaves, weighed and placed in 100 cm^3 of sterile physiological salt solution. All was thoroughly shaken for 30 minutes. Microbiological analysis was conducted using serial dilution method. The subsequent dilutions were sown on a recommended microbiological medium and incubated. The grown colonies with features characteristic for a given group were counted after the adequate culturing time. The test was performed in three replications and the results were averaged and expressed in c.f.u. (*colony forming units*) per bunch and per 100 g of radish.

Obtained results were elaborated statistically using one-way ANOVA and Tukey test. The test was applied when no equality between the averages was revealed. The analysis of variance was conducted on significance level $\alpha = 0.05$. Arithmetic mean, *standard deviation* (SD), range and variability coefficient (V %) were determined. The results were elaborated by means of Statistic 8.0 programme.

Results and discussion

Heavy metal contents in plants depend in the first place on the plant species but also on the cultivar, development stage, plant part and heavy metal content in soil, and finally on numerous environmental factors. In the presented research the analysis of variance revealed that the content of assessed metals was significantly diversified for the analysed radish part regarding zinc, chromium, lead and cadmium (Table 1).

Table 1

The content of heavy metals in radish [$mg \cdot kg^{-1}$ d.m.]

Heavy metal	Part of radisch	Range	Mean	SD	V %[1]	$LSD_{0.05}$
Zn	Leaves	36.40–635.70	101.79[b]	121.39	98	36.12
	Skin	43.20–360.25	81.17[ab]	60.29	74	
	Flesh	14.88–252.98	48.95[a]	50.20	96	
	Flesh + Skin	*35.11–300.07*	*77.30*[2]	*43.30*	*56*	
Cu	Leaves	2.30–58.30	5.84	9.93	84	ns[3]
	Skin	0–23.91	5.05	4.31	85	
	Flesh	1.20–4.13	2.26	0.80	36	
	Flesh + Skin	*2.39–28.20*	*4.38*	*3.45*	*79*	
Ni	Leaves	0–3.35	1.16	1.00	86	ns
	Skin	0–4.64	1.48	0.70	47	
	Flesh	0–6.04	1.04	0.55	53	
	Flesh + Skin	*0–4.67*	*0.80*	*0.56*	*70*	
Cr	Leaves	0.46–3.74	1.70[c]	0.89	52	0.31
	Skin	0.24–2.89	0.60[b]	0.51	86	
	Flesh	0–0.16	0.05[a]	0.03	70	
	Flesh + Skin	*0.24–0.26*	*0.78*	*0.43*	*55*	
Pb	Leaves	0.29–12.40	2.18[b]	2.13	98	0.64
	Skin	0.13–1.26	0.66[b]	0.29	45	
	Flesh	0–0.56	0.07[a]	0.12	69	
	Flesh + Skin	*0.31–0.52*	*0.97*	*0.74*	*76*	
Cd	Leaves	0.24–4.57	1.18[b]	0.93	79	0.49
	Skin	0–0.46	0.78[b]	1.35	45	
	Flesh	0.2–6.69	0.12[a]	0.11	89	
	Flesh + Skin	*0.11–2.93*	*0.70*	*0.54*	*77*	

[1] Variability coefficient; [2] weighted arithmetic mean; [3] non significant.

Analysing the highest mean contents of metals, the tested radish parts may be ordered decreasingly starting from the highest content: leaves > skin > flesh. A significant relationship was demonstrated only for nickel: skin > leaves > flesh. The differences between the lowest and highest metal content in the leaves ranged from 8 (Cr) to 33-fold (Cu), in the skin from 1 (Pb) to 17-fold (Zn) and in the flesh from 1 (Cu) to 33-fold (Cd). A high divergence of the assessed metal contents in the analysed radish parts was corroborated by the calculated values of variability coefficients, which for Zn, Cu, Ni and Pb were the highest in the leaves, while for Cr and Cd in the flesh (Table 1).

Depending on the tested part, the range of heavy metal contents in radish fluctuated from 14.88 to 635.70 mg Zn; from 0 to 58.30 mg Cu; from 0 to 6.04 mg Ni; from 0 to 3.74 mg Cr; from 0 to 12.40 mg Pb and from 0 to 5.57 mg Cd $\cdot$ kg^{-1} d.m. (Table 1). In comparison with the skin, radish leaves contained on average one time higher amounts of zinc, copper and cadmium, almost three times higher of chromium and over three times bigger Pb quantities. On the other hand, in comparison with radish flesh over once bigger quantities of nickel, twice bigger of zinc and copper, 36-fold higher amounts of lead and almost 10-fold higher quantities of cadmium were assessed in radish leaves (Table 1).

Mobility of metals in radish was determined using translocation index (T_I). The parameter was calculated as a ratio of metal contents in leaves and skin to their contents in flesh (Fig. 1).

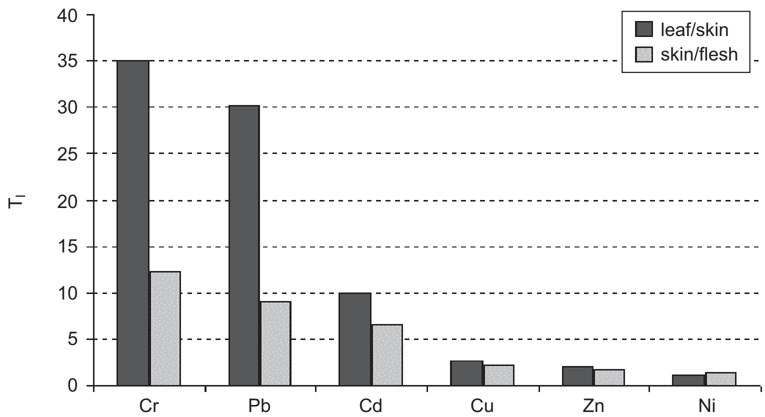

Fig. 1. Translocation coefficient of heavy metals in radish

The analysis of T_I value shows that radish nodosities (edible parts, flesh and skin) accumulated the greatest amounts of nickel, zinc and copper, slightly less cadmium and the least quantities of lead and chromium.

An interesting supplement to the above given data may be correlation coefficients computed between the contents of individual metals in radish depending on its analysed part (Table 2). As can be seen the highest number of correlations between the analysed metals was revealed in radish flesh. A significantly positive correlation was noted in flesh for Zn and Cr, Pb and Cd and subsequently for Cu and Cr and Pb, and also between Cr and Pb and for Cu and Cd. Investigations on the correlations between the contents of the analysed elements in radish skin showed a positive relationship for Zn and Ni and for Cr and Ni (Table 2). In leaves a significantly positive correlation was revealed for Cd and Zn and for Cr and Pb contents (Table 2). It should be noticed that in all three analysed plant parts a significantly positive correlation was noted between Pb and Cr, however it was the strongest in the flesh $r = 0.78$ ($p \leq 0.001$), weaker in the skin $r = 0.52$ ($p \leq 0.01$) and the weakest in the leaves $r = 0.37$ ($p \leq 0.05$).

Table 2

Values of simple correlation coefficients between the content of heavy metals in the analyzed part of the radish

Heavy metal	Zn	Cu	Cr	Ni	Pb
Leaves					
Cu	–0.06				
Cr	–0.33	0.02			
Ni	0.13	0.04	0.26		
Pb	–0.10	–0.03	0.37*	0.30	
Cd	0.40*	–0.07	–0.14	0.66***	0.06
Flesh					
Cu	0.21				
Cr	0.62***	0.41*			
Ni	0.62***	–0.13	0.15		
Pb	0.55**	0.5**	0.78***	–0.01	
Cd	0.44*	0.07	0.14	0.61***	0.11
Skin					
Cu					
Cr	–0.20	0.06			
Ni	0.73**	–0.10	–0.09		
Pb	0.36	0.16	0.52**	0.44*	
Cd	0.15	–0.04	–0.05	0.16	0.06

Significant at: *** $p \leq 0.001$, ** $p \leq 0.01$, * $p \leq 0.05$.

The assessment of the sanitary state of radish was presented in Table 3.

Table 3

Sanitary condition of radish

Parameter	*Coli* count		*Salmonella*		*Clostridium perfringens*	
	[cfu · bunch^{-1}]	[cfu · 100 g^{-1}]	[cfu · bunch^{-1}]	[cfu · 100 g^{-1}]	[cfu · bunch^{-1}]	[cfu · 100 g^{-1}]
Mean	135	80	6	3	1	1
Range	20–1300	11–664	20–120	17–61	10–20	6–10
% instances	36		10		2	

Presence of *Coli* and *Clostridium perfringens* bacteria evidences faecal contamination of not long ago. Presented research demonstrated that 64 % of radish samples (edible part flesh and skin) were clean of *Coli* bacteria, whereas 36 % were contaminated. On the other hand, spore forming bacteria *Clostridium perfringens* were found only in 2 % of samples and *Salmonella* in 10 % of samples (Table 3). The data demonstrate a good sanitary and hygienic state of the analysed radish purchased on the open air markets in Krakow.

A considerable percent of vegetables consumed by city dwellers originates from allotment gardens and plantations located in the suburban agricultural and horticultural holdings [10, 11]. It should be emphasized that vegetables belong to plants which very easily accumulate in their tissues excessive quantities of heavy metals, which change their chemical composition [4, 12]. Therefore, when cultivated in the regions subjected to city and industrial pollution they may not meet the quality criteria with respect to individual metals. Moreover, as reported by Filipek-Mazur et al [13] heavy metals, particularly high concentrations of lead and cadmium in plants for consumption, cause worsening of their quality and value as food. Norms of heavy metal contents in plants as suggested by Kabata-Pendias et al [14] admit consumption of plants with the following contents of heavy metals: ≤ 50 mg Zn; ≤ 20 mg Cu; ≤ 10 mg Ni; ≤ 1 mg Pb and ≤ 0.15 mg Cd $\cdot$ kg^{-1} d.m. Radish (edible part flesh and skin) assessment according to these norms shows that almost 40 % of the samples did not meet quality requirements of their usability for consumption due to excessive zinc concentrations whereas 65 % of samples because of cadmium content. Moreover, lead and cadmium contents in radish (edible part + skin) were assessed on the basis of European Commission Decree No. 1881/2006 of 19 December 2000 stating the highest admissible levels of some pollutants in foodstuffs [15]. According to the above-mentioned decree, the admissible content of lead and cadmium in root and stem vegetables should not exceed 0.1 mg Pb and 0.1 mg Cd $\cdot$ kg^{-1} f.m. Assuming a 10 % water content in the nodosities it corresponds to 1.0 mg Pb and Cd $\cdot$ kg^{-1} d.m. The studies demonstrated that none of the analysed radish samples exceeded the admissible lead level, whereas almost 10 % of radish samples revealed exceeded admissible cadmium level.

In conversion to 1 kg of fresh mass the studied radish contained in its nodosities (edible parts, flesh and skin) on average 7.73 mg Zn, 0.44 mg Cu, 0.08 mg Ni and Cr, 0.1 mg Pb and 0.07 mg Cd $\cdot$ kg^{-1} f.m. In the research conducted by Grembecka et al [3] radish originating from retail shops in the Gdansk region revealed almost thrice lower chromium content (0.03 mg $\cdot$ kg^{-1} f.m.) and 8-fold lower nickel content (0.01 mg $\cdot$ kg^{-1} f.m.) in comparison with radish bought on Krakow open air markets. The research of Tyksinski and Kurdubska [4] on cadmium and lead accumulation by radish indicated that increased doses of these metals in the substratum correspond to their elevated concentrations in radish nodosities. Moreover, the same authors revealed higher accumulation of cadmium and lead in plants cultivated in autumn than in spring. Average content of cadmium and lead in radish nodosities in autumn cultivation ranged from 0.96 to 11.57 mg Cd and from 4.49 to 20.83 mg Pb $\cdot$ kg^{-1} d.m., while in the spring cultivation metal concentrations were as follows: from 1.21 to 18.09 mg Cd and from 5.41 to 18.79 mg Pb $\cdot$ kg^{-1} d.m. [4]. High ability of radish to accumulate heavy metals is also testified by the fact that the above-mentioned authors registered high contents of cadmium and lead in radish cultivated in control combinations containing natural amounts of the metals. Jasiewicz [12] found that radish grown in soil containing 1.02 mg Cd $\cdot$ kg^{-1} d.m. accumulates in its nodosities 2.0 mg Cd $\cdot$ kg^{-1} d.m. Jurkowska et al [16] obtained similar results. The authors revealed that oil radish, spinach and fodder beet grown in the soil with natural cadmium and lead contents accumulated in their edible parts amounts of Cd and Pb which made them unsuitable for consumption [16].

The above quoted literature indicates that even at natural metal contents in the substratum, the admissible metal concentrations in vegetables are exceeded. It testifies a considerable ability of vegetables to accumulate heavy metals. The fact was confirmed by the results obtained by Curylo and Jasiewicz [17] who conducted research on carrot and celery and by Gaweda [18] in her studies on lettuce and radish.

Urbanized areas cover the city centre zones but also the peripheral parts of the cities. Increased content of heavy metals in soils is one of the pollution indicators in these areas [19]. Moreover, higher plants cultivated in the regions subjected to urban and industrial pollution may accumulate heavy metals even to the level hazardous to human health without showing any symptoms of toxicity [20–22]. According to the research of Rogoz and Opozda-Zuchmanska [21] zinc concentrations were exceeded in vegetables cultivated in the Krakow area and in the north-western regions, ie near Olkusz, Wadowice, Tyniec and Krzeszowice. Elevated cadmium concentrations in plants and soils in the north-western and north-eastern parts of Krakow in effect of dust emission from various industries were registered also by Gorlach and Gambus [23].

In conclusion excessive amounts of heavy metals and microbiological contamination occurring in foods of plant origin may cause various diseases. The sources of heavy metals may be both natural and anthropogenic. In Poland the arrangement of natural and anthropogenic factors may sometimes favour the uptake of greater metal amounts and their accumulation in usable plant parts [2, 24]. Moreover, as reported by numerous authors, plants growing in the regions under the influence of large industrial plants reveal higher contents of heavy metals than in agricultural areas [25].

Conclusions

1. The metals contents in radish ranged, depending on the analyzed part, from 14.88 to 635.70 mg Zn; from 0 to 58.30 mg Cu; from 0 to 6.04 mg Ni; from 0 to 3.74 mg Cr, from 0 to 12.40 mg Pb and from 0 to 5.57 mg Cd $\cdot$ kg^{-1} d.m.

2. The highest concentrations of zinc, copper, chromium, lead and cadmium were registered in leaves, lower in the skin and the lowest in radish flesh.

3. Radish did not fulfil consumption norms for zinc (40 % of analyzed samples) and cadmium (65 % of analyzed samples).

4. The sanitary state assessment of radish revealed *Coli* bacteria in 36 % of samples, *Salmonella* in 10 % of samples and *Clostridium perfringens* in 2 % of samples.

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ZAWARTOŚĆ METALI CIĘŻKICH I STAN SANITARNY JAKO OCENA JAKOŚCI RZODKIEWKI (*Raphanus sativum* L.)

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Abstrakt: Celem badań była ocena jakości zdrowotnej rzodkiewki pochodzącej z placów targowych Krakowa. Jakość zdrowotną oceniono na podstawie analizy zawartości i rozmieszczenia metali ciężkich w rzodkiewce, jak i jej stanu sanitarnego. Zawartość metali ciężkich (Zn, Cu, Cd, Pb, Cr, Ni) w miąższu, skórce i liściach rzodkiewki oznaczono metodą ICP-EAS po suchej mineralizacji i roztworzeniu popiołu w HNO_3. Analizy mikrobiologiczne obejmowały oznaczenie liczebność: bakterii z grupy coli, bakterii z rodzaju *Salmonella* oraz beztlenowych bakterii przetrwalnikujących *Clostridium perfringens*.

Zawartość metali ciężkich w badanej rzodkiewce wahała się w szerokim zakresie w zależności od analizowanej części. Największą zawartość cynku, miedzi, chromu, ołowiu i kadmu stwierdzono w liściach, mniejszą w skórce, a najmniejszą w miąższu rzodkiewki. Badana rzodkiewka nie spełniała norm konsumpcyjnych pod względem zawartości cynku i kadmu. Obecności bakterii grupy coli stwierdzono w 36 % próbkach, bakterii rodzaju *Salmonella* w 10 %, a *Clostridium perfringens* tylko w 2 %.

Słowa kluczowe: metale ciężkie, stan sanitarny, rzodkiewka