

# <sup>137</sup>Cs UPTAKE AND TRANSLOCATION IN LEAFY VEGETABLE: A STUDY WITH *Lactuca sativa* L. GROWN UNDER HYDROPONIC CONDITIONS

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**Abstract:** A hydroponic study involving lettuce plants (*Lactuca sativa* L.) as a leafy vegetable was conducted to evaluate the <sup>137</sup>Cs uptake and translocation in plant tissues in dependence on the presence or absence of K<sup>+</sup> or/and NH<sub>4</sub><sup>+</sup> ions in cultivation media according to Hoagland (HM) during 8 d plants growth under hydroponic conditions. Significant increase of the <sup>137</sup>Cs<sup>+</sup> uptake by lettuce plants and the decrease of <sup>137</sup>Cs<sup>+</sup> translocation efficiency from roots to leaves were observed in 50 % HM deficient in K<sup>+</sup> and NH<sub>4</sub><sup>+</sup> ions. Speciation analysis using Visual MINTEQ program showed that at micromolar concentration of CsCl (5 μmol/dm<sup>3</sup>) in 50 % HM at pH 6.0 and 25 °C, cesium was occurred practically only in the free cationic Cs<sup>+</sup> form – 98.8 %, with minor proportions of other cesium species: CsCl – 0.4 %, CsNO<sub>3</sub> – 0.4 %, and CsSO<sub>4</sub><sup>-</sup> – 0.4 %. Surplus of Cl<sup>-</sup>, NO<sub>3</sub><sup>-</sup> and SO<sub>4</sub><sup>2-</sup> ions in HM causes the increase of proportions of the cesium species CsCl, CsNO<sub>3</sub> and CsSO<sub>4</sub><sup>-</sup>, respectively at the expense of bioavailable Cs<sup>+</sup> form. Radiocesium <sup>137</sup>Cs taken up *via* roots was removed from lettuce leaves with high efficiency by boiling in diluted NaCl solution. At ambient temperature the extraction of <sup>137</sup>Cs with diluted acetic acid was concentration and time dependent process, and was succeeded by leakage of tissue components absorbing at 260 nm. These findings are important for the risk assessment of radiocesium entry into the food chain *via* contaminated leafy vegetable.

**Key words:** <sup>137</sup>Cs, root uptake, translocation, *Lactuca sativa*, speciation, food chain

## 1. Introduction

Contamination of the environment has become a serious problem all over the world. In contrast to heavy metals, which can present in high toxic levels in all parts of the environment, radionuclides are generally presented in low concentration, but from radiological unacceptable impact on live organisms point of view in non-negligible quantities. Especially, the presence of artificial radionuclides in the environment poses a serious risk to human health through the food chain. The artificial sources of radionuclides in the environment represent: testing of nuclear weapons (GABRIELI *et al.*, 2011), nuclear waste disposal (GRAMBOW, 2008), accidents resulting from the nuclear power generation as well as manipulation with nuclear fuel (HU *et al.*, 2010). In the case of the Chernobyl accident in 1986, the radionuclides released from the reactor that caused exposure of individuals were mainly radioiodine and radiocesium (UNSCEAR, 2000). These nuclides can be transferred to humans rapidly from the air *via* leafy vegetables, milk and other products of the food chain (ingestion pathway) (TSCHIERSCH *et al.*, 2009).

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From the radiological point of view, the releases of  $^{134}\text{Cs}$  ( $\tau = 2.06$  y) and  $^{137}\text{Cs}$  ( $\tau = 30.2$  y), estimated to have been 54 and 85 PBq, respectively, are the most important to consider (SOUDEK *et al.*, 2004). The accident of Fukushima nuclear power plant (NPP) on 11 March 2011 that resulted from the earthquake and tsunami contaminated large areas with radionuclides consisting primarily of  $^{131}\text{I}$ ,  $^{134}\text{Cs}$ , and  $^{137}\text{Cs}$  (MEXT, 2012). As reported by the Japanese Government to the IAEA on June 2011, the total amount of radioactivity released into the atmosphere might have been around  $1.5 \times 10^{17}$  Bq of  $^{131}\text{I}$  and  $1.5 \times 10^{17}$  Bq of  $^{137}\text{Cs}$ , and the total radioactivity discharged into the sea might have been around  $4.7 \times 10^{15}$  Bq (RJG, 2011). BAEZA *et al.* (2012) assume that released radioactivity correspond to approximately 6 % ( $^{131}\text{I}$ ) and 42 % ( $^{137}\text{Cs}$ ) of the radioactivity released into the environment after the nuclear reactor accident at Chernobyl.

Radionuclides released into the environment under normal or accidental functioning of the nuclear facilities, can contaminate plants *via* root uptake, direct deposition onto above-ground parts of plant or resuspension of contaminated soil particles. In the case of a nuclear accident occurring during the growing season, dry or/and wet deposition of aerosols on cultivated lands are the major and most direct steps in the food chain contamination (ANSPAUGH *et al.*, 2002). However, the release of new contamination still persists even after several years. Also, climate/environmental changes e.g. global warming (DOWDALL *et al.*, 2008) can play a possible role in distribution of radionuclides in soils as well as in soil-to-plant transfer processes. Following a nuclear accident during the growing period, long-lived radionuclides, compared to the time elapsed between contamination and harvest, could constitute a potential hazard to human when present as residues in consumed agricultural products. Ingestion of contaminated agricultural foodstuffs can constitute the major exposure pathway for the contribution to the total dose to human beings (MADOZ-ESCANDE *et al.*, 2004). It is therefore essential to study radionuclides uptake and translocation into edible parts of plants (mainly vegetables) as well as the effect of culinary preparation on radionuclide ingestion risks (see e.g. ADRIANO *et al.*, 2000). Contamination of the edible parts of „leafy vegetables“ (salad, for example) can comparatively occur throughout the growing period and contamination of ground vegetables, such as radishes, generally occurs through translocation from the contaminated aerial parts and root transfer from contaminated soil (MADOZ-ESCANDE *et al.*, 2004).

The bioassay using *L. sativa* L. (lettuce) studies is considered a simple, quick and sensitive way of evaluating potential environmental risk (GOPALAN, 1999) and can be successfully applied in soil with a high concentration of heavy metals of natural and anthropogenic origin (FARRÉ and BARCELÓ, 2003; ESCOTO *et al.*, 2007; BAGUR-GONZÁLEZ *et al.*, 2011). World production of lettuce in the year 2009 was approximately 24 million metric tons, with a cultivation area of one million hectares (USDA, 2011). However, the significant production of lettuce is realized under hydroponic conditions. Hydroponic cultivation has been reported not only to be associated to higher production yields but also to allow better control and standardization of the cultivation process, thus reducing overall production costs (DOMINGUES *et al.*, 2012).

Our previous papers reported bioaccumulation and distribution of cesium ( $^{137}\text{Cs}$ ), cobalt ( $^{60}\text{Co}$ ), cadmium ( $^{109}\text{Cd}$ ) and zinc ( $^{65}\text{Zn}$ ) in tissues of tobacco (*Nicotiana tabacum* L.) (VRTOCH *et al.*, 2007; HORNÍK *et al.*, 2007; HORNÍK *et al.*, 2009; GULDANOVÁ *et al.*, 2010), celery (*Apium graveolens* L.) (HORNÍK *et al.*, 2007) and sunflower (*Helianthus annuus* L.) (HORNÍK *et al.*, 2005; HORNÍK *et al.*, 2007). Also, the foliar uptake and translocation of zinc ( $^{65}\text{Zn}$ ) in tobacco (*N. tabacum* L.) and hop (*Humulus lupulus* L.) plants we studied (PIPIŠKA *et al.*, 2008; MAREŠOVÁ *et al.*, 2012). This paper deals with  $^{137}\text{Cs}$  uptake and translocation in lettuce (*Lactuca sativa* L.) plants as a model of leafy vegetable contaminated with radiocesium and behaviour of radiocesium after treatment of lettuce leaves in the context of radiocesium releasing from contaminated leaf tissues.

## 2. Materials and methods

### 2.1. Plant material

Seeds of lettuce (*Lactuca sativa* L. var. *capitata* L.) obtained from Seva Seed, Ltd., CR were germinated and grown in pots with commercial garden substrate (AGRO CS Slovakia, SR) and illuminated by natural sunlight. The plants were periodically watered with tap water. After 3-4 weeks of pre-cultivation seedlings were gently removed from the substrate, roots were thoroughly washed by deionized water for removing of substrate particles and used in hydroponic experiments.

### 2.2. Hydroponic experiments

Selected lettuce plants with more than 10 leaves (height approx. 10 cm, 3 g wet weight) from the pre-cultivation phase were transferred into series of 250 cm<sup>3</sup> Erlenmeyer flask containing 150 cm<sup>3</sup> of 50 % Hoagland medium (HOAGLAND, 1920) spiked with  $^{133}\text{CsCl}$  and  $^{137}\text{CsCl}$  as a tracer. Cultivation medium was adjusted with 1 M NaOH to the value pH 6.0.

The composition of the full strength (100 %) Hoagland medium was (mmol/dm<sup>3</sup>): MgSO<sub>4</sub>·7H<sub>2</sub>O – 1.5; KNO<sub>3</sub> – 4.0; CaCl<sub>2</sub> – 4.0; NaH<sub>2</sub>PO<sub>4</sub>·2H<sub>2</sub>O – 2.1; Na<sub>2</sub>HPO<sub>4</sub>·12H<sub>2</sub>O – 0.13; FeSO<sub>4</sub>·7H<sub>2</sub>O – 6.4·10<sup>-2</sup>; NaNO<sub>3</sub> – 4.0; NH<sub>4</sub>Cl – 4.0; NH<sub>4</sub>NO<sub>3</sub> – 2.0; H<sub>3</sub>BO<sub>3</sub> – 0.14; Na<sub>2</sub>MoO<sub>4</sub>·2H<sub>2</sub>O – 2.5·10<sup>-4</sup>; MnSO<sub>4</sub>·5H<sub>2</sub>O – 2.1·10<sup>-2</sup>; ZnSO<sub>4</sub>·7H<sub>2</sub>O – 2.3·10<sup>-3</sup>; CuSO<sub>4</sub>·5H<sub>2</sub>O – 3.2·10<sup>-3</sup>.

All flasks were covered with black foil to protect plant roots against the lights and plants were cultivated under hydroponic conditions in triplicate series at artificial illumination (4 000 lx) and 16 h light / 8 dark photoperiod. In time intervals aliquot samples of cultivation media were taken, remaining  $^{137}\text{Cs}$  radioactivity was measured by gamma-spectrometry and subsequently samples were returned back into the medium. At the same time the reduction of cultivation medium volume in flasks caused by transpiration activity of plants was recorded and the difference was compensated by glass balls addition due to keeping of roots submersion in media. At the end of experiments, lettuce plants were removed from cultivation media, roots

were carefully washed in deionized water and incorporated  $^{137}\text{Cs}$  radioactivity in roots and aboveground parts of plants was analysed. Dry weight of roots and leaves was estimated after 48 h drying at 60 °C.

Growth value (GV) for lettuce plants was estimated according to the equation (1):

$$GV = \frac{m_t - m_0}{m_0} \quad (1)$$

where  $m_t$  or  $m_0$  are fresh weight of plants at the start or end of experiments, respectively.

### 2.3. Extraction of $^{137}\text{Cs}$ from lettuce leaf tissues

One-step extraction of  $^{137}\text{Cs}$  from leaves of lettuce plants obtained from hydroponic experiments in 50 % HM spiked with  $^{137}\text{CsCl}$  was performed by diluted acetic acid under occasional shaking at [biomass] : [extractant] ratio 1 : 50 and  $23 \pm 2$  °C.

### 2.4. Radiometric analysis

Two gamma-spectrometric scintillation detectors 54BP54/2-X and 76BP76/3 with well type crystal NaI(Tl) (Scionix, NL) and data processing software ScintiVision-32 (Ortec, USA) were used for determination of  $^{137}\text{Cs}$  in cultivation medium and individual parts of lettuce plants. For energy and efficiency calibration a library of radionuclides was built by selecting characteristic  $\gamma$ -ray peaks (88.04 keV for  $^{109}\text{Cd}$ , 661.66 keV for  $^{137}\text{Cs}$ , 834.81 keV for  $^{54}\text{Mn}$  and 1115.52 keV for  $^{65}\text{Zn}$ ). Standardized solution of  $^{137}\text{Cs}$  in the form of  $^{137}\text{CsCl}$  ( $5.723 \text{ MBq/cm}^3$ ,  $20 \text{ mg/dm}^3$   $^{137}\text{CsCl}$  in  $3 \text{ g/dm}^3$  HCl) was provided from Czech Metrological Institute (Prague, CR).

### 2.5. Speciation modelling

The proportion of individual Cs ionic forms in cultivation medium was predicted by speciation modelling software Visual MINTEQ (ver. 3.0) obtained from GUSTAFSSON (2010) in consideration of total salt concentration in medium, pH value, ionic strength and temperature.

### 2.6. Statistical analysis

All analytical determinations were performed in triplicate. Statistical significance of differences in calculated values of  $^{137}\text{Cs}$  distribution and translocation in lettuce plant tissues on the basis of different cultivation conditions were evaluated by multiple range test to ascertain differences between individual groups. The level of significance was  $p = 0.05$  in all cases. Origin 8.5 Pro (OriginLab Corp., USA) and STATGRAPHICS Centurion ver. 15 (StatPoint, Inc., USA) were used for graphing and statistical analyses, respectively.

### 3. Results and discussion

#### 3.1. The effect of $K^+$ and $NH_4^+$ ions on $^{137}Cs$ uptake

Uptake and translocation of  $^{137}Cs$  in lettuce plants (*L. sativa* L.) were studied under hydroponic conditions. These conditions on the one hand imitate in some details conditions of soil solution and on the other hand represent one way of commercial production of lettuce. Thus, from the point of view of soil contamination with radiocesium the mentioned arrangement of experiments describes only  $^{137}Cs$  transport in sequence soil solution – plant roots – aboveground parts of plant without characterisation of sorption-desorption processes between soil particles and soil solution. It is generally known, that  $^{137}Cs$  shows high affinity to sorption onto clay minerals (aluminosilicates). From this reason the radiocesium binding in mentioned soil components is a limiting factor, which determines the behaviour, bioavailability and transport of Cs into the environment. In generally, these processes are also affected by chemical speciation of analysed metal or radionuclide in given environment e.g. the portion of individual ionic form of metal or radionuclide in soil solution.

Table 1. Chemical speciation of cesium in complete 50 % Hoagland medium (HM), in HM deficient in  $K^+$  and  $NH_4^+$  ions and in HM with surplus of  $Cl^-$ ,  $NO_3^-$  and  $SO_4^{2-}$  anions. Calculated by Visual MINTEQ ver. 3.0 for pH 6.0 and 25 °C and  $C_0 = 5 \mu\text{mol}/\text{dm}^3$  CsCl.

Hoagland medium	Molar fraction [x].100			
	$Cs^+$	$CsCl$	$CsNO_3$	$CsSO_4^-$
HM complete	98.8	0.4	0.4	0.4
HM deficient for $K^+$	99.0	0.4	0.2	0.4
HM deficient for $NH_4^+$	99.0	0.3	0.3	0.4
HM deficient for $K^+$ and $NH_4^+$	99.1	0.2	0.2	0.5
Surplus of $Cl^-$ anions*				
10-times (60.005 mmol/dm <sup>3</sup> )	96.2	3.1	0.3	0.4
100-times (600.005 mmol/dm <sup>3</sup> )	80.4	<b>19.2</b>	0.2	0.2
Surplus of $NO_3^-$ anions*				
10-times (50.000 mmol/dm <sup>3</sup> )	95.8	0.3	3.5	0.4
100-times (500.000 mmol/dm <sup>3</sup> )	78.5	0.2	<b>21.1</b>	0.2
Surplus of $SO_4^{2-}$ anions*				
10-times (7.950 mmol/dm <sup>3</sup> )	95.3	0.3	0.4	4.0
100-times (79.500 mmol/dm <sup>3</sup> )	78.0	0.2	0.2	<b>21.6</b>

\* Concentration of anions in 50 % HM (mmol/dm<sup>3</sup>):  $Cl^-$  – 6.005;  $NO_3^-$  – 5.000;  $SO_4^{2-}$  – 0.795.

In this context the speciation modelling program Visual MINTEQ for prediction of individual ionic forms of Cs in Hoagland media (HM) was used. We found that in 50 % HM containing  $5 \mu\text{mol}/\text{dm}^3$  CsCl the cesium was occurred mainly in the free cationic form  $Cs^+$  (98.8 %) with minor presence of CsCl (0.4 %),  $CsNO_3$

(0.4 %) and  $\text{CsSO}_4^-$  (0.4 %) forms at pH 6.0 and 25 °C (Tab. 1). In the case cultivation media without  $\text{K}^+$  ( $\text{KNO}_3$  as source),  $\text{NH}_4^+$  ( $\text{NH}_4\text{Cl}$  and  $\text{NH}_4\text{NO}_3$  as source) or without both cations the slight increase of free  $\text{Cs}^+$  (up to 99.1 %) was observed. On the basis of  $\text{CsCl}$ ,  $\text{CsNO}_3$  and  $\text{CsSO}_4^-$  existence in 50 % HM under given conditions, it can be concluded that in the decrease or increase of  $\text{Cs}^+$  cations portion in media the concentration of  $\text{Cl}^-$ ,  $\text{NO}_3^-$  or  $\text{SO}_4^{2-}$  anions will play important role. In Tab. 1, it is evident that if we increase 10-times or 100-times the concentration of mentioned anions the  $\text{Cs}^+$  portion in 50 % HM will significantly decrease. This phenomenon can be expected also in real soil conditions and soil solution.

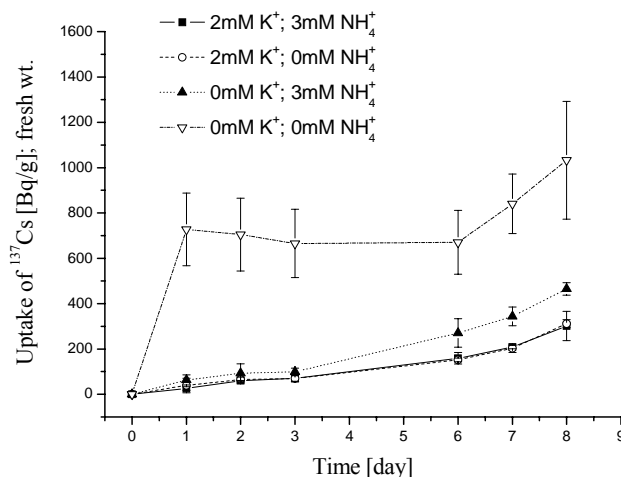


Fig. 1. The effect of absence or presence of  $\text{K}^+$  and  $\text{NH}_4^+$  ions in cultivation media on cesium uptake by root system of lettuce plants (*L. sativa* L.) during 8 d hydroponic cultivation in 50 % Hoagland medium (HM) spiked with  $5 \mu\text{mol}/\text{dm}^3$   $\text{CsCl}$  ( $36.4 \text{ kBq}/\text{dm}^3$   $^{137}\text{CsCl}$ ), pH 6.0 at photoperiod 16 h light / 8 h dark (4 000 lx) and  $25 \pm 2$  °C. Error bars represent standard deviation of the mean ( $n = 3$ ). The initial average fresh weight (fresh wt.) of lettuce plants was 2.68 g and the average increase of plants weight during experiments was  $GV = 1.26 \pm 0.15$  (SD;  $n = 12$  – the mean of values obtained at all cultivation conditions).

Fig. 1 depicts Cs uptake by root system of lettuce plants (*L. sativa* L.) during 8 days hydroponic cultivation in the complete 50 % HM spiked with  $5 \mu\text{mol}/\text{dm}^3$   $\text{CsCl}$  ( $36.4 \text{ kBq}/\text{dm}^3$   $^{137}\text{CsCl}$ ) and in 50 % HM without  $\text{K}^+$ ,  $\text{NH}_4^+$  or without both ions. The choice of plant cultivation time length was adequate to evaluation of  $^{137}\text{Cs}$  uptake and translocation into lettuce plant tissues. SABBARESE *et al.* (2002) observed that in his time the maximum radionuclide  $^{60}\text{Co}$  or  $^{137}\text{Cs}$  transport was reached in the case of lettuce plants. We found that the significant increase of  $^{137}\text{Cs}^+$  uptake by lettuce plants was observed in 50 % HM fully deficient in both  $\text{K}^+$  and  $\text{NH}_4^+$  ions. In media deficient in  $\text{K}^+$  or  $\text{NH}_4^+$  ions the significant increase of Cs uptake, but in less extent, was shown for  $\text{K}^+$  ions. Also, we observed that in complete media or media without  $\text{K}^+$  or  $\text{NH}_4^+$  ions the Cs uptake by roots of lettuce plants showed time linear dependence. However, in the case of media deficient in both  $\text{K}^+$  and  $\text{NH}_4^+$  ions can be seen three phases: i) the first phase characterized by rapid Cs uptake (after 24 h more than 60 % of the total Cs was uptake); ii) the second equilibrium phase and iii) the third phase

of gradually increasing in Cs uptake from solution of 50 % HM. All these differences in Cs uptake at different composition of 50 % HM can not be explained on the basis of differences in chemical speciation of Cs in media (see previous paragraph). Results of authors SOUDEK *et al.* (2004; 2006) show, that the concentrations of  $K^+$  and  $NH_4^+$  ions in media play a decisive role as competitive ions (inhibitors) in Cs uptake by plants. CHOU *et al.* (2005) found that the transfer of  $^{137}Cs$  from contaminated soil to lettuce plants (*L. sativa* L.) was significantly diminished in the case of amended soil with 100 ppm KCl. These facts are crucial from the point of view of possible risks of  $^{137}Cs$  transfer from contaminated environment to food chain e.g. through leafy vegetables. Thus, in the case of the environment (soil or water) deficient mainly in  $K^+$  ions, there is a high risk of plants contamination with  $^{137}Cs$ . On the other hand, the environment rich in  $K^+$  or  $NH_4^+$  ions inhibit the transfer of  $^{137}Cs$  into plants by competitive relationships between these macroelements and  $^{137}Cs^+$  cations. In this context, in Table 2 we present determined specific radioactivity  $^{137}Cs$  (Bq/g; fresh wt.) in lettuce plants at different  $[Cs^+] : [K^+]$ ,  $[Cs^+] : [NH_4^+]$  or  $[Cs^+] : [K^+ + NH_4^+]$  molar concentration ratios.

Table 2. Content of  $^{137}Cs$  [Bq/g] (fresh wt.) in lettuce plants (*L. sativa* L.) after 2 and 8 d growth in 50 % HM spiked with  $5 \mu mol/dm^3$  CsCl ( $36.4 \text{ kBq/dm}^3$   $^{137}CsCl$ ) at different  $[Cs^+] : [K^+]$ ,  $[Cs^+] : [NH_4^+]$  or  $[Cs^+] : [K^+ + NH_4^+]$  molar concentration ratios.

$C_0$ [mmol/dm <sup>3</sup> ]		Molar ratio			Radioactivity [Bq/g]	
$K^+$	$NH_4^+$	$[Cs^+] : [K^+]$	$[Cs^+] : [NH_4^+]$	$[Cs^+] : [K^+ + NH_4^+]$	2 d	8 d
0.0	0.0	-	-	-	704.7	1032.7
0.0	3.0	-	1 : 600	-	92.7	465.4
2.0	0.0	1 : 400	-	-	65.4	311.8
2.0	3.0	-	-	1 : 1 000	60.5	301.6

It is generally known that the uptake of  $Cs^+$  is operated mainly *via* two transport pathways on plant root cell membranes, namely  $K^+$  transporters (high-affinity mechanism) or  $K^+$  channels (low-affinity mechanism). However, the physiological role of  $Cs^+$  in plant nutrition until now is no known (MARSCHNER, 1995; WHITE and BROADLEY, 2000). The change in  $^{137}Cs$  uptake at an external K concentration in nutrient solution around  $0.25 \text{ mmol/dm}^3$ , coincides with the concentration range at which the K uptake systems switch between a high-affinity mechanism and a low-affinity mechanism (FU and LUAN, 1998; WAEGENEERS *et al.*, 2001).

It can be expected that differences in K uptake rates in the genotype level result in different K depletion and hence different radiocesium uptake rates. WAEGENEERS *et al.* (2001) found that these differences are more pronounced in soils with a low K availability and reported that high radiocesium uptake occurs (i) if the plant species has a high intrinsic Cs uptake rate at low K concentrations (e.g. barley, lettuce, bentgrass), (ii) if the species have a high biomass production, resulting in a large depletion of K in the bulk soil solution (e.g. ryegrass), or (iii) if the species have a high K uptake rate per unit root surface, resulting in a high rhizospheric K depletion (e.g. lettuce).

### 3.2. The effect of $K^+$ and $NH_4^+$ ions on $^{137}Cs$ translocation

From the point of view of risks of toxic metals or radionuclides transfer into the food chain it is important that given metals or radionuclides will not be accumulated mainly in edible parts of plants. In the Fig. 2 is presented the effect of  $K^+$  and  $NH_4^+$  ions deficiency in cultivation media on distribution of  $^{137}Cs$  in root and leaves of lettuce plants after 8 days hydroponic cultivation. We found that the deficiency in  $K^+$  and  $NH_4^+$  ions caused a decreasing the accumulation of  $^{137}Cs$  in leaves from the total accumulated radioactivity in lettuce plants in comparing with roots (significant differences are shown in Fig. 2 at the  $p = 0.05$  level based on multiple range test). In the complete 50 % HM (control) the portion of  $^{137}Cs$  radioactivity in lettuce leaves represents more than 70 % of the total accumulated amount of  $^{137}Cs$ . The absence of  $K^+$  and  $NH_4^+$  ions this amount of  $^{137}Cs$  in leaves decreased on the value less than 40 %. Thus, it can be concluded that the presence of  $K^+$  and  $NH_4^+$  ions in media increase the Cs transport from roots to leaves of lettuce plants whereby this effect is additive.

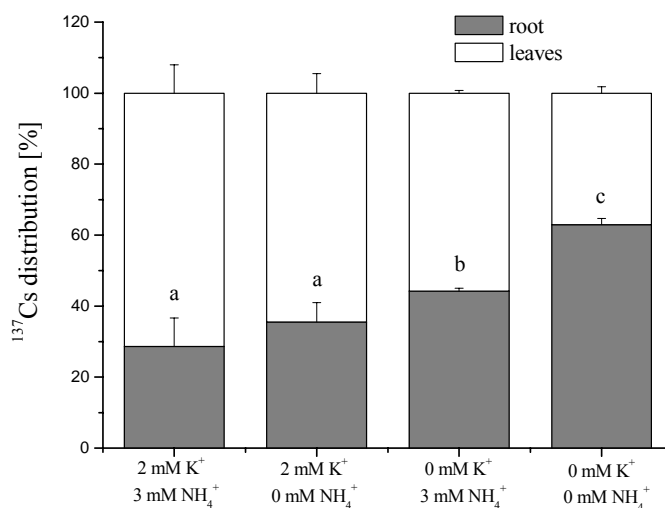


Fig. 2. Distribution of cesium in roots and leaves of lettuce plants grown 8 d at the absence or presence of  $K^+$  and  $NH_4^+$  ions in HM media. For details see legend in the Fig. 1. Error bars represent standard deviation of the mean ( $n = 3$ ). Means with the same letter at columns are not significantly different at the  $p = 0.05$  level based on multiple range test.

For evaluation of  $^{137}Cs$  mobility and translocation in conductive tissues of lettuce plants we analysed the distribution of radiocesium on the basis of the ratio of concentration in edible part of plants  $[Cs]_{leaves}$  to concentration in root system of plants  $[Cs]_{root}$ . Also, as can be seen in the Fig. 3, in the case of lettuce plants cultivation in 50 % HM fully deficient in both  $K^+$  and  $NH_4^+$  ions was reached 4.5-times lower value of the concentration ratio  $[^{137}Cs]_{leaves} : [^{137}Cs]_{root}$  (Bq/g : Bq/g; dry wt.) than in control experiments with the value  $[^{137}Cs]_{leaves} : [^{137}Cs]_{root} = 0.73$ .



Our previous results (GULDANOVÁ *et al.*, 2010) obtained for tobacco plants cultivated under hydroponic conditions showed that the concentration ratio  $[Cs]_{shoot} : [Cs]_{root}$  increased with increasing HM concentration from the value 0.10 to the value 0.85, particularly at 50 % and 100 % HM, when concentration of monovalent ions ( $K^+$  and  $NH_4^+$ ;  $[K^+] : [NH_4^+] = 2 : 3$ ) were 5 mmol/dm<sup>3</sup> and 10 mM, respectively. Similar effect was also observed with sunflower hydroponics, when shoot-to-root specific <sup>137</sup>Cs radioactivity ratio (Bq/g : Bq/g; wet wt.) increased with increasing concentration of HM from the value 0.10 to the value 0.69 (HORNÍK *et al.*, 2005). CHOU *et al.* (2005) found that in the contaminated soil with <sup>137</sup>Cs the addition of 100 ppm KCl caused the increase of the transfer factor of <sup>137</sup>Cs for the aboveground and the decrease for the underground parts of cabbage, spinach, lettuce, radish, rape and clover plants cultured in contaminated soil during 7, 30 or 50 days.

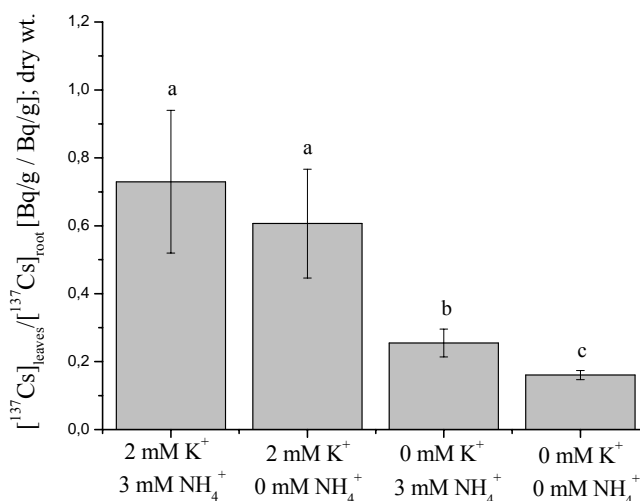


Fig. 3. The effect of  $K^+$  and  $NH_4^+$  ions on the  $[Cs]_{leaves} / [Cs]_{root}$  concentration ratio of lettuce plants after 8 d growth in 50 % HM. For details see legend in the Fig. 1. Error bars represent standard deviation of the mean ( $n = 3$ ). Means with the same letter at columns are not significantly different at the  $p = 0.05$  level based on multiple range test.

In the case of contaminated plants is very important to know not only the distribution of radioactivity in roots and aboveground parts, but also the distribution in individual segments of aboveground parts (old and young leaves, fruits, stems). In this term we analysed the specific radioactivity  $A_S$  (Bq/g; dry wt.) for <sup>137</sup>Cs in individual leaves of lettuce plants (head and wrapper leaves) cultivated in complete 50 % HM or media without  $K^+$  or/and  $NH_4^+$  ions. We found that the highest specific radioactivity in individual lettuce leaves was in the case of plants cultivated in 50 % HM deficient in  $K^+$  and  $NH_4^+$  ions and  $A_S$  decreased in the order of plants cultivated in: HM deficient in  $K^+$  and  $NH_4^+$  (7 950 Bq/g) > HM deficient in  $K^+$  (4 160 Bq/g) > HM deficient in  $NH_4^+$  (3 940 Bq/g) > complete HM (3 220 Bq/g). Also we determined that values of the specific radioactivity  $A_S$  in all

leaves within the analysed plant were not significantly differed. From this result, it can be concluded that the uptake of  $^{137}\text{Cs}$  in individual leaves of lettuce plant will not be dependent on the development stage or localization of the leaf.

### 3.3. Releasing of $^{137}\text{Cs}$ from lettuce leaves

Many papers deal with radionuclide releasing from contaminated vegetables and other foods during culinary preparation (see e.g. ADRIANO *et al.*, 2000) or during decontamination by washing of the plant matter after dry deposition of radionuclides (see e.g. TSCHIERSCHE *et al.*, 2009).

In this part of the paper we evaluate the effect of lettuce leaves treatment on the residual concentration of radiocesium in leaves. All the  $^{137}\text{Cs}$  radioactivity in experiments entered the plant *via* the roots from  $^{137}\text{Cs}$  spiked HM and was localized within the plant tissues.

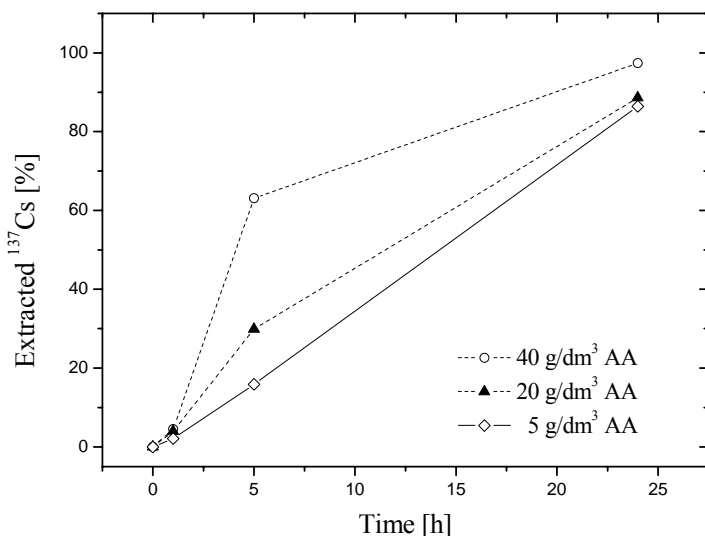


Fig. 4. One-step extraction of  $^{137}\text{Cs}$  from leaves of lettuce plants grown in 50 % HM spiked with  $^{137}\text{CsCl}$ . Extraction with diluted acetic acid (AA) at [biomass] : [extractant] ratio 1 : 50 and  $23 \pm 2$  °C. Leaves of lettuce with specific radioactivity 370 Bq/g (fresh. wt.) were obtained from hydroponic experiments described in Fig. 1.

Radiocesium was removable from lettuce leaf biomass with high efficiency by boiling in diluted NaCl solutions (data not shown). The efficiency of extraction under mild conditions such as by extraction with diluted organic acids at ambient temperature was low and time dependent.

Negligible amounts of  $^{137}\text{Cs}$  were removed from the lettuce leaves after 1 h contact with diluted acetic acid and for higher efficiency of the process several hours contact times were necessary (Fig. 4). The highest efficiency of  $^{137}\text{Cs}$  removal was obtained only after 24 hour treatment with diluted acetic acid. However, at these conditions leaf

tissue was significantly damaged, decolorized and into the extraction media were released cell components absorbed at 260 – 280 nm region, typical for nucleic acid and proteins (Fig. 5).

The question of Cs speciation and distribution in green plants is not completely solved. Prevailing part of cesium will exist in a form of free  $\text{Cs}^+$  ion and the main barrier of the  $\text{Cs}^+$  ions translocation are diffuse barriers of the plant cells. In other biosystems e.g. in basidiomycetes cesium can form complexes with organic ligands such as badione or norbadione structurally related to the larger pigment family of pulvinic acids (DESAGE-EL MURR *et al.*, 2006).

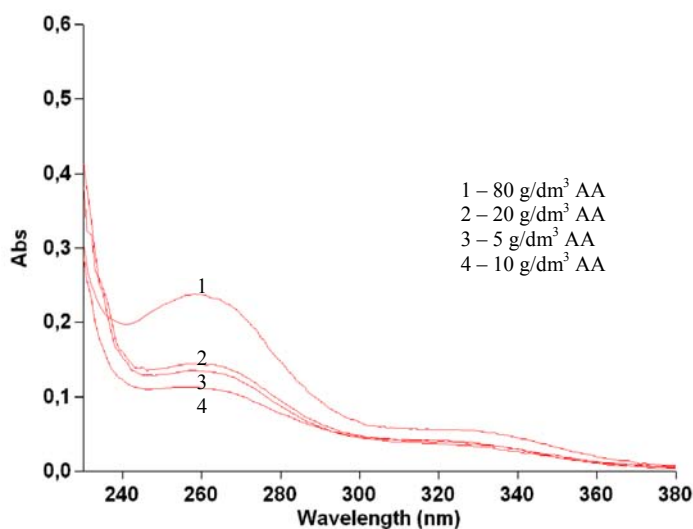


Fig. 5. UV-VIS spectrum of diluted acetic acid solutions (AA) after 24 h extraction of  $^{137}\text{Cs}$  from lettuce leaves at  $23 \pm 2$  °C. For details see the legend at Fig. 4.

#### 4. Conclusions

The results from hydroponic cultivation of lettuce plants (*L. sativa* L.) indicate that the uptake and translocation of  $^{137}\text{Cs}$  in plant tissues are significantly affected by presence and concentration of  $\text{K}^+$  and  $\text{NH}_4^+$  ions. We found that the significant increase of  $^{137}\text{Cs}^+$  uptake by lettuce plants was observed in cultivation media deficient in both  $\text{K}^+$  and  $\text{NH}_4^+$  ions. On the other hand, the absence of mentioned ions in media caused a decreasing of  $^{137}\text{Cs}$  translocation from roots to leaves of lettuce plants. From the point of view of risks of toxic metals or radionuclides transfer into the food chain we also evaluate the effect of lettuce leaves treatment on the residual concentration of radiocesium in leaves. Radiocesium was removable from lettuce leaf biomass with high efficiency by boiling in diluted NaCl solutions and in the case of mild conditions with diluted organic acids at ambient temperature low and time dependent releasing of  $^{137}\text{Cs}$  from contaminated lettuce leaves was observed.

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