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Biometric traits of onion (*Allium cepa* L.) exposed to ^{137}Cs and ^{243}Am under hydroponic cultivation

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ABSTRACT

To elucidate the features of bioaccumulation and phytotoxic effects of long-lived artificial radionuclides, a hydroponic experiment was carried out with the cultivation of onion (*Allium cepa* L.) in low-mineralized solutions spiked with ^{137}Cs (250 kBq L^{-1}) or ^{243}Am (9 kBq L^{-1}). After the 27-day growth period, ≈70% of ^{137}Cs and ≈14% of ^{243}Am were transferred from the solutions to onion biomass with transfer factor values ≈ 400 and ≈ 80, respectively. Since the bioaccumulation of both radionuclides mainly took place in the roots of onion (77% ^{137}Cs and 93% ^{243}Am of the total amount in biomass), edible organs – bulbs and leaves – were protected to some extent from radioactive contamination. At the same time, the incorporation of the radionuclides into the root tissues caused certain changes in their biometric (geometric and mass) traits, which were more pronounced under the ^{243}Am-treatment of onion. Exposure to ^{243}Am significantly reduced the number, length, and total surface area of onion roots by 1.3–2.6 times. Under the influence of ^{137}Cs, the dry-matter content in roots decreased by 1.3 times with a corresponding increase in the degree of hydration of the root tissues. On the whole, the data obtained revealed the specific features of ^{137}Cs and ^{243}Am behaviour in "hydroponic solution – plant" system and suggested that biometric traits of onion roots could be appropriate indicators of phyto(radio)toxicity.

1. Introduction

Since the 1940–50s, mankind has been dealing with artificial radionuclides, which began to spread widely in the environment as a result of nuclear weapons tests in the atmosphere, the activities of the nuclear fuel cycle enterprises, and especially in cases of major accidents with the subsequent fallout of nuclear fission products to the Earth's surface (Beresford et al., 2016; IAEA, 2004; Shaw, 2007). Total discharge of radionuclides during the accidents in Kyshtym (1957, USSR) was estimated as $7.4 \cdot 10^{16}$ Bq, in Windscale/Sellafield (1957, UK) – $7.7 \cdot 10^{14}$ Bq, in Chernobyl (1986, USSR) – $5.3 \cdot 10^{18}$ Bq, in Goiania (1987, Brazil) – $5.1 \cdot 10^{13}$ Bq; and during the Fukushima Dai-Ichi accidental release of radionuclides was estimated to be approximately one-tenth of the Chernobyl emission (Alexakhin, 2009; IAEA, 2004, 2015). As a result, the Northern Hemisphere is currently contaminated with long-lived radionuclides at the global level.

Nowadays and over the next few decades, ^{137}Cs will continue to be the most significant ambient radioactive pollutant marking the areas of bomb-derived and post-accidental radioactive fallout (UNSCEAR, 2008). In particular, the total amount of Chernobyl ^{137}Cs deposited in European terrestrial ecosystems was estimated at 64 PBq; and the area where the fallout exceeded 40 kBq m^{-2} was about $200 \cdot 10^3$ km^2, including $3.6 \cdot 10^3$ km^2 of the territory of the former USSR (Belarus, Ukraine, the European part of Russia) with soil contamination >1480 kBq m^{-2} (Atlas of caesium deposition …, 1998). The external dose rate in these territories is mainly controlled by ^{137}Cs activity. Radiocaesium is a long-lived ($T_{1/2}$ 30.17 y) artificial radioisotope of naturally occurring monovalent alkali metal ^{133}Cs, which undergoes β-decay followed by γ-emission at a high energy level of 661.7 keV. Due to the strong irreversible fixation of ^{137}Cs by clay and mica minerals of the soil (Durrant et al., 2018; Kruglov et al., 2008) and its corresponding insignificant downward migration in Chernobyl-affected lands the radionuclide is still mainly located in the top horizon (humus or ploughed one) (Burger and Lichtscheidl, 2018; Paramonova et al.,

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2015). The mechanisms of ^{137}Cs uptake by plant roots are supposed to be comparable with the absorption of essential K^+- and NH_4^+-ions, therefore, a rather high bioavailability of radiocaesium is generally presumed (White and Broadley, 2000; Zhu and Smolders, 2000).

Transuranic elements are found in the environment in small amounts and mainly dispersed globally through the nuclear weapons test fallout; they are only marginally concentrated in the immediate vicinity around the test sites and accident zones (Atlas of caesium deposition …, 1998; Kashparov et al., 2003). Actinides have almost no effect on the ambient external dose rate, but cause concern because of their high ecotoxicity even at trace concentrations (Kabata-Pendias, 2010; Malátová and Bečková, 2014). The unique radionuclide is ^{241}Am ($T_{1/2}$ 432.2 y), which pool is replenished and reaches a maximum 72 years after the incident of environmental pollution as a result of the decay of the parent ^{241}Pu ($T_{1/2}$ 14.4 y). Modern ^{241}Am soil contamination in the exclusion Chernobyl zone (Ukraine) is estimated to be in the range of 0.2–6.3 kBq kg^{-1} (Horemans et al., 2018), in the area of the former Semipalatinsk Test Site (Kazakhstan) – 0.3–4.9 kBq kg^{-1} (Kozhakhanov et al., 2014); but the maximum activity of ^{241}Am in terrestrial ecosystems of Europe is expected only in ~2060–70 s at a total activity of 0.077 PBq (UNSCEAR, 2008). In the end, after ≈300 years, ^{241}Am will be the most significant residual radionuclide in post-Chernobyl landscapes. ^{241}Am is polyvalent (III-VI) metal with α-decay mode accompanied by relatively mild γ-emission at 59.5 keV energy line. There is one more long-lived radioisotope of americium – ^{243}Am, which has still less abundance in the environment but is often used for radiochemical applications and analysis. ^{243}Am is characterized by α-decay and γ-emission at 74.7 keV, and its geochemical properties, including phytotoxicity, are similar to those of ^{241}Am (Malátová and Bečková, 2014). In acidic soils and waters, predominant Am(III) ion is readily chelated and enters into the inner part of organic substances; at neutral-alkaline pH conditions its behaviour is additionally controlled by carbonate and hydroxyl complexes (Kabata-Pendias, 2010). Due to the strong affinity to high molecular organic substances $^{241(243)}$Am is predominantly immobilized in humus horizons of soils (Kozhakhanov et al., 2014; Lehto et al., 2013). Root uptake of $^{241(243)}$Am is two or three orders of magnitude lower than that of ^{137}Cs, but Am is presumed to be one of the most bioavailable amongst transuranic element (Kabata-Pendias, 2010; Whicker et al., 1999).

Being accumulated in topsoil, *i.e.* in the rooting zone of plants, ^{137}Cs and $^{241(243)}$Am are inevitably involved in the process of biological migration and potentially can cause hazardous effects, especially in a short food chain "arable soil – crop – (domestic animal) – human". Severe environmental risks can arise even at the first stage of the radionuclides entering the "soil-plant" system. In this regard, the phytotoxicity of the radionuclides at the biochemical, physiological and morphological levels, as well as the degree of bioaccumulation of ^{137}Cs and $^{241(243)}$Am in plants, are of serious concern.

The radioactive isotopes of caesium and especially americium have phytotoxic properties. In the experiment with cultivation of thale cress (*Arabidopsis thaliana*) in a presence of a rather low activity of ^{134}Cs Sahr et al. (2005) noted the gradual disruption of gene-expression profile in roots and general oxidative stress leading to a significant reduction in the growth rate of plants. Bioaccumulation of ^{137}Cs in tobacco (*Nicotiana tabacum*) was additionally accompanied by a slowdown in growth and necrosis or die-back of leaves at concentrations of CsCl over 0.2 mM (Guldanová et al., 2010). Among the biochemical characteristics of plants, exposure to ^{134}Cs decreased the total chlorophyll content in the pokeweed (*Phytolacca americana*) and amaranth (*Amaranthus cruentus*) (Tang et al., 2011). The naturally occurring stable ^{133}Cs, which is non-essential heavy metal, also has potential phytotoxic properties: at concentrations above 200 μM L^{-1} it induced a considerable reduction in the number of germinated seeds and elongation of lettuce (*Lactuca sativa*) seedlings (De Medici et al., 2019), loss of length and dry weight of roots and leaves of plantain (*Plantago major*) (Burger et al., 2019a); reduced root elongation in broad bean (*Vicia faba*) (Zhang et al., 2020), root biomass and shoot length of wheatgrass (*Agropyron cristatum*), cheatgrass (*Bromus tectorum*) (Cook et al., 2009), sweet basil (*Ocimum basilicum*) (Ko et al., 2018) and napiergrass (*Pennisetum purpureum*) (Kang et al., 2012). It is believed that caesium can inhibit plant growth due to impaired potassium uptake by roots (Burger et al., 2019a, 2019b; Le Lay et al., 2006), modification of the jasmonate pathway (Adams et al., 2013), and a decrease in intracellular inorganic phosphates (Kamel et al., 2007) or an increase in the content of soluble sugars (Le Lay et al., 2006).

Phytotoxicity of americium isotopes is supposed notably strong even at trace concentrations (Malátová and Bečková, 2014). For instance, in hydroponically cultivated *A. thaliana*, a noticeable decrease in root mass was revealed under the influence of both high (50 kBq L^{-1}) and low (0.05 kBq L^{-1}) specific activity of ^{241}Am in solutions (Biermans et al., 2015).

Stress sensitivity/tolerance of plants is apparently largely determined by the biological features of crops and varies widely. In particular, the approximately 5-50-fold difference in intensity of ^{137}Cs transfer from the same growth medium into a crop was observed depending on plant species and varieties (Staunton et al., 2003; Waegeneers et al., 2001; White et al., 2003). Onion (*Allium cepa* L.) is thought to have a high ^{137}Cs accumulation capacity compared to other plant species (Bystrzejewska-Piotrowska and Urban, 2004). Besides, it is recommended as a suitable monocotyledonous crop very sensitive to acute and chronic contaminations of various natures (EPA, 1996; OECD, 2003). From an applied perspective, onion bulbs and leaves are an important part of human consumption worldwide with an annual production of 93.2 million tons (in 2016), with constantly growing onion harvesting area (Hanci, 2018). Consequently, in the event of intensive bioaccumulation of ^{137}Cs and/or $^{241(243)}$Am in edible parts of onions, radionuclides can quickly be incorporated in the human food chain, which will create a toxicological risk not only for plants but also for the population.

The present study was conducted under the conditions of a hydroponic experiment, which provided the highest bioavailability of radionuclides for plants and, accordingly, triggered the most dramatic onion responses to the exposure of radiocaesium and americium. By this means it was supposed to elucidate: (i) quantitative estimation of bioaccumulation degree of ^{137}Cs and ^{243}Am in onion; (ii) patterns of the radionuclides translocation and distribution between roots, bulbs and leaves of onion; (iii) possible peculiar features of morphological (biometric) response of onion to ^{137}Cs and ^{243}Am treatment. The first and second items were important in terms of predicting the radionuclides root uptake from low-mineralized solutions and their distribution between edible and inedible parts of onion. The latter item has been chosen due to the relative simplicity of plant biometric indicators measurement along with the integral nature of the morphological changes, reflecting molecular, cellular and physiological mechanisms of plant growth and development.

2. Material and methods

2.1. Plant material and design of the hydroponic experiment

The hydroponic experiment was conducted with bulbs of onion Stuttgarter Riesen cultivar commercially gained at a local seed market. Dormant bulbs without roots and visible signs of damage, approximately the same size and weight (1.2–1.4 cm, 3 ± 0.3 g) were selected for the experiment.

Before planting, the tops of the bulbs were slightly cut to initiate the growth process. The bulbs were weighed and placed in special plastic 2 L containers, each with 10 holes, filled with settling tap water spiked with either: a) ^{137}CsCl with a specific activity of 250 kBq L^{-1} (^{137}Cs-treatment) or b) ^{243}AmNO$_3$ with a specific activity of 9 kBq L^{-1} (^{243}Am-treatment). The cultivation of onion in tap water without adding radionuclides served as a control. All variants of the experiment were carried out in ten biological replicates. Each bulb was planted into an individual funnel to prevent evaporation of the solution from the uncovered surface.

Activity concentrations of the radionuclides in solutions were adjusted to subtoxic levels, approximately equal to ¼ of the quantities that clearly affect the germination of seedlings, in accordance with the "Root Elongation Toxicity Test" (EPA, 1996). Tap water was used as a growth medium instead of a standard nutrient solution to reduce ions competition/complexation by virtue of low mineralization and neutral pH: K 4, Na 23, Ca 54, Mg 14, Fe < 0.05, Mn 0.017, Al 0.02, Cu 0.007, Pb < 0.001, Sr 0.15, Zn 0.054, As <0.005, Cd < 0.0005, NH_4^+ 0.15, NO_3^- 3, NO_2^- 0.005, HCO_3^- 183, CO_3^{2-} <6, SO_4^{2-} 5, Cl^- 26, F^- <0.3 mg L^{-1}; pH 7,1 (the data are placed at disposal by the analytical laboratory of Chemistry Faculty of Lomonosov Moscow State University). The pH of all solutions was controlled during the experiment and maintained at a neutral range of 6–7 to create the best conditions for plant growth and to prevent complexation of ^{243}Am ions with carbonate and hydroxyl compounds.

Cultivation of onion was conducted in a growth chamber for 27 days until the plants reached the mature stage of "leaves development". The plants were grown using supplementary lighting generated by a 15-W LED-based phytolamp with 12 h exposure during the day with photon flux density 250 μmol m^{-2} s^{-1} and 12 h photoperiod; at 23/18 °C day/night temperature; and at air humidity of 45–60%. Hydroponic solutions were constantly aerated by an air pump. The number of leaves and their length were daily recorded.

2.2. Plant sampling and primary processing of biometric traits

After 27 days of vegetation the onion plants were removed from the containers, rinsed 3 times with distilled water, thoroughly washed with running tap water for 1 min, rinsed in 40% ethanol solution for 3 min to remove possible external contamination with insoluble compounds from the root surface and finally rinsed again 3 times with distilled water. Thereafter each individual plant sample was dried with filter paper, separated into roots, bulbs, and leaves, and weighed to determine its fresh biomass. Simultaneously roots and leaves of each plant were photographed with resolution of 150 dpi for determination of their morphological parameters.

Then roots, bulbs and leaves of two randomly-systematically selected plants grown under ^{137}Cs- and ^{243}Am-treatments were prepared for the further digital autoradiography study. Remaining plants were integrated into a uniform sample, dried at the temperature of 80 °C in an oven for 24 h, weighed, and then ground using a mortar and pestle for homogenization of plant material, and prepared for the further γ-spectrometric analysis.

Digital photos of roots and leaves were processed using special free access programs ARIA (ComPM Lab @ Iowa State University, USA), RootSystemAnalyser 1.0 (D Leinter, A Schnepf) for roots, and LAMINA (Umea University, Sweden) and LeafJ (Maloof lab @ University of California Davis, USA) plugin of ImageJ for leaves. Based on the data from this processing of digital images, as well as taking into account fresh and dry biomass, the biometric characteristics of onion's grown in control and exposed to ^{137}Cs and ^{243}Am treatments were calculated.

2.3. Laboratory analyses

Digital autoradiography of the plant organs – roots, bulbs and leaves – was performed using PerkinElmer Cyclone device (USA). The exposure time was differently selected for the onion's organs; on average, it was 3–4 h for bulbs and 23–24 h for roots and leaves.

Spectrometric measurements were made using a GR 3818 semiconductor γ-spectrometer with a Canberra high-purity (HPGe) detector (USA). The efficiency coefficients were 0.015 for ^{137}Cs and 0.13 for ^{243}Am. The specific activity of the radionuclides in prepared onion samples was measured in the standard geometry of the Petri dish (diameter 3.5 cm). Energy spectra measurements were accounted for 1200 s, and ^{137}Cs and ^{243}Am activities were calculated from the net full energy peaks 661.7 keV and 74.7 keV, respectively, using the "Genie 2000" spectrometric analysis software package. The analytical error of γ-spectrometry did not exceed 2%.

2.4. Calculation of the radionuclides root uptake and translocation

To assess the intensity of ^{137}Cs and ^{243}Am transfer from hydroponic solution into onion the values of total transfer factor (TF_{tot}) were calculated as (1):

$$TF_{tot}^{Cs-137}(Am-243) = A^{Cs-137}(Am-243)_{plant} \Big/ A^{Cs-137}(Am-243) \quad (1)$$

where $A^{Cs-137(Am-243)}{}_{plant}$ is specific activity of ^{137}Cs (or ^{243}Am) in dry biomass of plant (kBq g^{-1}), and $A^{Cs-137(Am-243)}{}_{solution}$ is specific activity of ^{137}Cs (or ^{243}Am) in solutions (kBq mL^{-1}), correspondently.

Special transfer factors from hydroponic solution into onion organs (TF_{leaves}, TF_{bulbs}, and TF_{roots}) for the onion organs – leaves, bulbs, roots – were calculated as (2):

$$TF_{leaves(bulbs,roots)}^{Cs-137(Am-243)} = A_{leaves(bulbs,roots)}^{Cs-137(Am-243)} \Big/ A^{Cs-137(Am-243)}\text{solution} \quad (2)$$

where $A^{Cs-137(Am-243)}{}_{leaves\ (bulbs,\ roots)}$ is specific activity of ^{137}Cs (or ^{243}Am) in dry biomass of the relevant onion organ (kBq g^{-1}).

To quantify the transfer of ^{137}Cs from roots to aerial parts the translocation factor (TLF) was calculated as (3):

$$TLF_{leaves(bulbs)}^{Cs-137(Am-243)} = A_{leaves(bulbs)}^{Cs-137(Am-243)} \Big/ A_{roots}^{Cs-137(Am-243)} \quad (3)$$

Overall root uptake (ORU) of the radionuclides was defined as a percentage of the total content of ^{137}Cs or ^{243}Am in onion plants from their initial amount in solutions (4):

$$ORU^{Cs-137(Am-243)} = \left(A^{Cs-137(Am-243)}\text{plant} \bullet TDB^{Cs-137(Am-243)}\right) \Big/ \left(A^{Cs-137(Am-243)}\text{solution} \bullet V_{solution}\right) \bullet 100 \quad (4)$$

where $TDB^{Cs-137(Am-243)}$ is dry biomass of onion exposed to ^{137}Cs and ^{243}Am treatments respectively (see also section 2.5); and $V_{solution}$ is the total volume of hydroponic solution used, equal to 2 L.

2.5. Analyses of biometric traits

The following biometric characteristics of onion were recorded (all – per one plant) (Garnier et al., 2001; Pérez-Harguindeguy et al., 2013):

- Number of leaves (LN);
- The longest leaf length (LL_{max}), *i.e.* plant height –a distance between dry onion flakes and the tip of the longest leaf, cm;
- Leaf elongation relative rate (LERR) – increment of the longest leaf length to its length in the beginning of the observation period, %;
- Leaf area (LA) – total surface area of leaves, cm^2;
- Number of roots (RN);
- The longest root length (RL_{max}) – a distance between basal plate and the tip of the longest root, cm;
- Root average length (RL_{avg}) – mean value of roots length, cm;
- Root overall length (RL_{sum}) – sum of the lengths of all roots, cm;

- Root diameter (RD) – mean diameter value of all roots measured by the length of each, cm;
- Root area (RA) – total surface area of all roots, cm^2;
- Leaf fresh biomass (LFB), bulb fresh biomass (BFB), root fresh biomass (RFB), total fresh biomass (TFB), g;
- Leaf dry biomass (LDB), bulb dry biomass (BDB), root dry biomass (RDB), total dry biomass (TDB), mg;
- Leaf dry-matter content (LDMC), bulb dry-matter content (BDMC), root dry-matter content (RDMC), total dry-matter content (TDMC) – dry biomass of onion or its organ, divided by corresponding fresh biomass, $mg\ g^{-1}$;
- Specific leaf area (SLA) – total surface area of all leaves, divided by their dry biomass, $cm^2\ mg^{-1}$;
- Specific root area (SRA) – the ratio of total surface area of all roots to dry biomass of roots, $cm^2\ mg^{-1}$;
- Specific root length (SRL) – the ratio of sum length of roots to dry biomass of roots, $mm\ mg^{-1}$.

2.6. Statistical analyses

The obtained data were analyzed by application of Microsoft Excel 2003 (Microsoft Cooperation, USA) for the basic descriptive statistics and Statistica 8.0 (Statsoft, USA) for the one-way analysis of variance (ANOVA) with post-hoc Tukey's test to determine the significance of the difference between the mean values. Statistical significance was assumed at $\alpha < 0.05$.

3. Results and discussion

3.1. Growth dynamics and viability signs of onion under ^{137}Cs and ^{243}Am exposure

Observation of the onion growth dynamics through the control of elongation the first (later on – the longest) leaf and the number of leaves per plant demonstrated only small and statistically insignificant suppression under ^{137}Cs- and ^{243}Am-treatments. The first green shoot was observed on the 6th day after planting for all plus/minus radionuclide treatments. Although, in the control, somewhat more uniform germination was noticed (half of the bulbs versus single exemplars under ^{137}Cs and ^{243}Am exposure). Complete germination of the onion bulbs occurred on the 10th day in the control and on the 12th day under hydroponic cultivation with the radionuclides when the phenological stage of the clearly visible (>3 cm) 1st leaf was reached by all plants.

The onion growth dynamics occurred as damped exponential (Fig. 1), which is typical for the bulb vegetables (Pérez-Harguindeguy et al., 2013). The leaf elongation parameters were similar in all treatments of the experiment. A very small (statistically insignificant) difference in the length of the longest leaf was found only during the 3rd week of cultivation, when the average L_{max} of onion exposed to the radionuclides was 10–25 mm shorter than in the control. In the same period, the number of leaves after treatment with ^{137}Cs and ^{243}Am was insignificantly higher than in the control (on average 2.4–2.5 versus 1.8 per plant, respectively). Similar effect of inhibition the napiergrass (*Pennisetum purpureum*) growth accompanied by multiplication of a number of stems under exposure to 300–3000 mM ^{133}Cs in hydroponic solutions was reported by Kang et al. (2012).

At the same time, at the 3rd week of onion cultivation, corresponding to the period of the most intensive shoots elongation, LERR values were slightly higher under exposure to the radionuclides: 183% when growing plants with ^{137}Cs, 177% with ^{243}Am, and 150% in the control. By the 21st day of the experiment, the difference between L_{max} in all treatments was smoothed out, and the rate of further growth by LERR index varied within the narrow limits of 29–31%.

Radionuclides seemingly have stronger or weaker root uptake degree and different phytotoxic effects at certain stages of vegetation. In support of this, for example, after γ-irradiation of *Arabidopsis*, expression of mismatch repair genes was less in 10- or 30-day-old plants compared to 20-day-old plants because they were in the process of transition to reproductive growth and had a special need for genome stability (Sidler et al., 2015). Also, with an equal external activity of ^{241}Am in solution, the roots of 14-day-old *Arabidopsis* had a 10-fold higher activity than 18-day-old ones (Biermans et al., 2015). In the current experiment, starting from the 21st day after planting on the phenological stage of a clearly visible 3rd leaf, the mean number of onion leaves and L_{max} values were levelled across all treatments.

Areas of the main accumulation of radionuclides in plants can be marked by corresponding signs of phytotoxicity or nutrient deficiency. For example, there was chlorosis occurred in the leaves of golden pothos

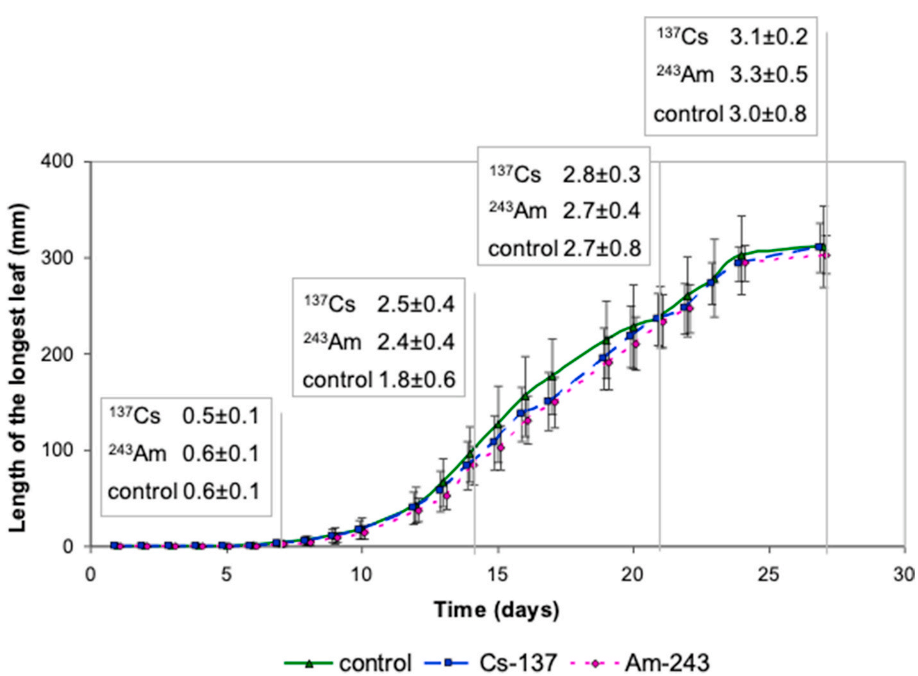

Fig. 1. Dynamics in the onion growth during hydroponic cultivation in the control conditions and under ^{137}Cs- and ^{243}Am-treatment. The data are mean ± CI (confidence interval) of the longest leaf length. Data in the highlighted boxes are the average number of leaves per plant ± CI at the end of the 1st, 2nd, 3rd, and 4th week of the cultivation, respectively. The differences between the longest leaf length and leaves number in both treatments and the control are statistically insignificant (for 10 biological replicates at $\alpha < 0.05$).

(*Epipremnum aureum*) exposed to 198 kBq L^{-1} of ^{137}Cs after 15 days of hydroponic cultivation (Kamel et al., 2007). In the present study, there were no visible signs of phytotoxicity in the aerial part of onion – chlorosis, brown points, leaf wilting – at least until the harvesting of onion after 27 days of cultivation in solutions with ^{137}Cs 250 and ^{243}Am 9 kBq L^{-1}. However, onion roots were partially characterised by darkening and point when exposed to ^{137}Cs and ^{243}Am (3 and 2 plants of 10, respectively). In addition, under the influence of ^{243} Am-containing solutions, a weak symptom of root gelatinization was observed.

3.2. Root uptake of ^{137}Cs and ^{243}Am from hydroponic solutions, root-to-shoot translocation and distribution between organs of onion

In general, there are no precise data on the uptake of radionuclides by roots of bulbous vegetables, and even the IAEA summary report "Handbook of Parameter Values for Predicting the Transport of Radionuclides in Terrestrial and Freshwater Environments" (IAEA, 2010) does not provide information on ^{137}Cs and/or ^{243}Am bioaccumulation in this group of plants. Only a few studies are known to evaluate the transfer of radionuclides to the onion. Thus, onion grown in the Chernobyl-affecting agrosystem of Poland was characterized by TF 2.9 (Bystrzejewska-Piotrowska and Urban, 2004), whereas the bulk of agricultural crops of temperate climate exhibited TF < 1 (IAEA, 2010). When Bystrzejewska-Piotrowska and Urban (2004) selected onion as a test-crop for a hydroponic experiment, they found still further root uptake of caesium and concluded that onion could be considered a potential phytoremediator for radioactively contaminated soils and waters.

In the present study, an intensive transfer of ^{137}Cs and ^{243}Am into onion biomass with TF_{tot} n · 10^1-10^2 values was noted, which was accompanied by considerable bioaccumulation of the radionuclides in onion tissues (Table 1). In doing so both radionuclides were presented in hydroponic solutions in ionic forms with high potential bioavailability, but $TF^{Cs-137}{}_{tot}$ was approximately 5 times more than $TF^{Am-243}{}_{tot}$. As for $^{137}Cs^+$, it is possible that the similarity of non-specific mechanisms of its ion transport with those for essential K^+ and NH_4^+ (Burger and Lichtscheidl, 2018; Hampton et al., 2005; White and Broadley, 2000; Zhu and Smolders, 2000) was beneficial for more intensive uptake by plant roots and further symplastic transport to shoots. Relating to $^{243}Am^{3+}$, there are no essential nutrients comparable with this radionuclide by the mechanism of ionic transport. In addition, a gradual decrease in its bioavailability from hydroponic solution could occur as a result of the radionuclide complexation with root exudates released during plant growth.

Actually, TF^{Cs-137} and TF^{Am-243} values might seem extremely high at first glance, but usually radionuclide transfer to aquatic plants in freshwater ecosystems, even taking into account partial immobilization of radionuclides by natural solid particles (suspensions and bottom sediments), is several orders of magnitude greater than in terrestrial ecosystems. To illustrate, reference TF^{Cs-137} values for edible aquatic plants obtained from the field measurements vary within the range 1.9–3.3 · 10^3, and TF^{Am-243} – 7.5 – 3.9 · 10^4, versus soil-to-cereals TF^{Cs-137} 2.0 · 10^{-4} – 9.0 · 10^{-3}, and TF^{Am-241} – 7.4 · 10^{-7} – 3.4 · 10^{-2} (IAEA, 2010). With a high level of radioactive contamination in natural water bodies within the 30-km Chernobyl exclusion zone, the highest TF^{Am-241} for aquatic plants reached 7500 (Gudkov et al., 2002).

In experimental conditions, it was found that ^{133}Cs was absorbed by wild calla (*Calla palustris*) from a hydroponic solution, 210 times more than from contaminated soil (Rinaldi et al., 2017). However, in general, the quantitative assessments of the values of $TF^{Cs-137(Cs-133)}$ and $TF^{Am-241(Am-243)}$ in "hydroponic medium-plant" systems vary greatly, mainly depending on the biological features of the test-crops and composition of the growth solution (Table 2).

In the present study, the evidence obtained on a rather high degree of ^{137}Cs and ^{243}Am root uptake by onion had demonstrated considerable bioaccumulation ability of the crop, but it should be mentioned that the conditions of the hydroponic experiment assumed the greatest bioavailability of both radionuclides, since low mineralized solutions with small amounts of K^+ and NH_4^+ and neutral pH provided depletion of the competitive ions and/or complexing agents. Such chemical composition of solutions essentially promote $^{137(133)}$Cs (De Medici et al., 2019; Genies et al., 2017; Guldanová et al., 2010; Šuňovská et al., 2012), as well as $^{241(243)}$Am (Whicker et al., 1999) root uptake by plants, however, only rarely occurs in natural conditions. In addition, radiocaesium consumption was discovered to increase significantly at the shoot growth stage (Marčiulioniene et al., 2008), which onion has reached in the current experiment.

Evaluation of the initial and final balance of the radionuclide in "hydroponic solution – plant biomass" showed substantial overall bioaccumulation of ^{137}Cs in onion after 27 days of cultivation. The obtained ORU^{Cs-137} value was equal to 69.8%, fitted neatly into the range of the same estimations of previous hydroponic studies with various experimental design, plant species and exposure time (Fig. 2). Of the other crops, onion displayed relatively elevated efficiency of ^{137}Cs root uptake from solutions, but even with beneficial experimental conditions, the levels of the radionuclide specific activity observed in plants corresponded only to a ^{137}Cs concentration of 3.1 · 10^{-2} mg kg^{-1}, which is much lower value of 1% dry weight, characterizing plant species with the ability for a hyperaccumulation (Baker and Brooks, 1989).

The migration efficiency of ^{243}Am in the "hydroponic solution – onion biomass" system was significantly reduced in comparison with ^{137}Cs: ORU^{Am-243} value did not exceed 14.3%.

The distribution of the radionuclides amongst onion's organs is the matter of particular interest since the bulbs and leaves are edible ones. The literature data on the distribution of $^{137(133)}$Cs over plant tissues are rather contradictory. Some authors postulated a higher specific activity of radiocaesium in roots (De Medici et al., 2019; Eapen et al., 2006; Kamel et al., 2007; Moogouei et al., 2017; Rinaldi et al., 2017; Singh et al., 2009; Staunton et al., 2003; Šuňovská et al., 2012; Wang et al., 2012); others confirmed an increased specific activity of ^{137}Cs in the aerial part (Brambilla et al., 2002; Fulekar et al., 2010; Kang et al., 2012; Wang et al., 2000). Meanwhile, the researchers sometimes pointed out the various ^{137}Cs distribution patterns between roots and shoots for different plant species (Paramonova et al., 2015; Soudek et al., 2006, 2004; Yasutaka et al., 2014). In the present study, the specific activity of ^{137}Cs was maximal in the roots, decreased by ~ 40 times in the bulbs, and again slightly rose in the leaves (≈6 times compared to bulbs).

According to existing data, $^{241(243)}$Am, like other transuranic and rare earth elements, should mainly accumulate in plant roots (Biermans et al., 2015, 2014; Duffa et al., 2002; Kabata-Pendias, 2010; Kozhakhanov et al., 2014). None of the researchers described a different distribution pattern of $^{241(243)}$Am in plant tissues. The results of the present

Table 1
Characteristics of ^{137}Cs and ^{243}Am accumulation in onion biomass under hydroponic cultivation.

Part of plant	^{137}Cs-treatment				^{243}Am-treatment			
	Activity, kBq g^{-1}	Amount per 1 plant, kBq	TF	TLF	Activity, kBq g^{-1}	Amount per 1 plant, kBq	TF	TLF
Leaves	46.8	6.7	189	0.15	0.002	0.0003	0.3	0.001
Bulbs	8.2	1.0	33	0.03	0.13	0.018	14.6	0.047
Roots	317	26.8	1282	–	2.88	0.245	313	–
Total biomass	98.8	34.6	399	–	0.76	0.264	82.2	–

Table 2
Transfer factors of ^{137}Cs and ^{243}Am in different experimental hydroponic conditions.

Ecotoxicant, activity or concentration	Hydroponic solution	Duration of exposure to ecotoxicants	Test-crop	Part of plants	TF	Reference
$^{133}CsCl$, 0.15–15 μM	Artificial groundwater: $Ca(NO_3)_2$ 0.7, $MgSO_4$ 0.5, KNO_3 1.5 mM L^{-1}; and Na_2HPO_4 1 μM L^{-1}	48 h	Indian mustard (*Brassica juncea*)	Plants	100–250	Salt et al. (1997)
^{137}Cs, 5 kBq L^{-1}	NH_4NO_3 4.24, KNO_3 0.14, $CaCl_2$ 0.25, $Ca(NO_3)_2$ 0.77, KH_2PO_4 0.5, K_2SO_4 1.27, $MgSO_4$ 0.46 mM L^{-1}; KOH and micronutrients	11–64 days	Wheat (*Triticum aestivum*)	Shoots	30–60	Smolders and Shaw (1995)
				Roots	60–140	
$^{133}CsCl$, 1 μM	KNO_3 1.2, $Ca(NO_3)_3$ 0.4, $NH_4H_2PO_4$ 0.1, $MgSO_4$ 0.2 mM L^{-1}; KCl 50, H_3BO_3 12.5, $NiSO_4$ 0.1, $ZnSO_4$ 1.0, $CuSO_4$ 0.5, $MnSO_4$ 1.0, H_2MoO_4 0.1, Fe^{3+}-EDDHA (for dicots) or Fe^{3+}-HEDTA (for grasses) 10, and Mes-Tris 1.0 μM L^{-1}	28 days	Cabbage (*B. oleracea* var. *capitata*)	Shoots	165	Lasat et al. (1997)
			Tepary bean (*Phaseolus acutifolius*)	– "–	143	– "–
			Indian mustard (*B. juncea*)	– "–	120	– "–
			Hairy vetch (*Vicia villosa*)	– "–	113	– "–
			Broccoli (*B. oleracea* var. *botrytis*)	– "–	105	– "–
			Cauliflower (*B. oleracea* var. *botrytis*)	– "–	105	– "–
			Kochia (*Kochia scoparia*)	– "–	60	– "–
			Reed canarygrass (*Phalaris arundinacea*)	– "–	45	– "–
			Colonial bentgrass (*Agrostis capillaris*)	– "–	45	– "–
			Red fescue (*Festuca rubra*)	– "–	38	– "–
$^{133}Cs_2SO_4$, 0.002–20 mM	Nitsch's nutrient solution: KNO_3 1.5, NH_4NO_3 0.72, $CaCl_2$ 1, $MgSO_4$ 0.5 mM L^{-1}, H_3BO_3 0.1, Na_2EDTA 0.2; and $FeSO_4$ 0.027, $MnSO_4$ 0.025, $ZnSO_4$ 1, $CuSO_4$ 1, Na_2MoO_4 1 μM L^{-1}	1 month	Broadleaf plantain (*Plantago major*)	Plants	0.8–6.8	Burger et al. (2019a)
$^{133}Cs_2SO_4$, 0.002–20 mM	– "–	7 weeks	Rockcress (*Arabidopsis halleri*)	– "–	0.8–16.5	Burger et al. (2019b)
^{137}Cs, 46–198 Bq mL^{-1}	KCl 2.012, $MgSO_4$ 0.997, $Ca(NO_3)_2\cdot 4H_2O$ 4.006, KH_2PO_4 0.45, H_3BO_3 0.011, $FeCl_3$ 0.004 mM L^{-1}; and $ZnSO_4 \cdot 7H_2O$ 2, $CuCl_2 \cdot .2H_2O$ 1, $Na_2MoO_4 \cdot 2H_2O$ 1, $MnCl_2 \cdot 4H_2O$ 1 μM L^{-1}	1–15 days	Golden pothos (*Epipremnum aureum*)	– "–	12.5–36.0	Kamel et al. (2007)
^{133}Cs (CsCl), 50–3000 μM	Hoagland's nutrient solution: $MgSO_4 \cdot 7H_2O$ 1.5, KNO_3 4.0, $CaCl_2$ 4.0, $NaH_2PO_4 \cdot 2H_2O$ 2.1, $Na_2HPO_4\cdot 12H_2O$ 0.13, $FeSO_4 \cdot 7H_2O$ 0.064, $NaNO_3$ 4.0, NH_4Cl 4.0, NH_4NO_3 2.0 H_3BO_3 0.14 mM L^{-1}, $MnSO_4\cdot 5H_2O$ 0.021; and $Na_2MoO_4\cdot 2H_2O$ 0.25, $ZnSO_4\cdot 7H_2O$ 2.3, $CuSO_4\cdot 5H_2O$ 3.2 μM L^{-1}	7 weeks	Napiergrass (*Pennisetum purpureum*)	– "–	0.7–1.5	Kang et al. (2012)
$^{133}CsCl$, 0.5–1 mM	tap water with the addition of K_2SO_4 0.5–10 mM L^{-1}	8 days	Wild calla (*Calla palustris*)	– "–	0.5–1.4	Komínková et al. (2018)
^{137}Cs, 1–10 MBq L^{-1}	distilled water	1–15 days	Siam weed (*Chromolaena odorata*)	– "–	0.5–0.9	Singh et al. (2009)
$^{241}AmCl_3$, 2.23 kBq L^{-1}	modified Hoagland's solution: KNO_3 1, $Ca(NO_3)_2$ 0.3, $MgSO_4$ 0.2, $NH_4H_2PO_4$ 0.1, $FeSO_4$ 1.62, Na_2EDTA 0.78, H_3BO_3 4.6, $MnCl_2$ 0.9, $CuSO_4$ 0.032, H_2MoO_4 0.055, and $ZnSO4 \cdot 7H_2O$ 0.077 mM L^{-1}	72 h	*Arabidopsis thaliana*	Shoots	13.5	Biermans et al. (2014)
				Roots	715	

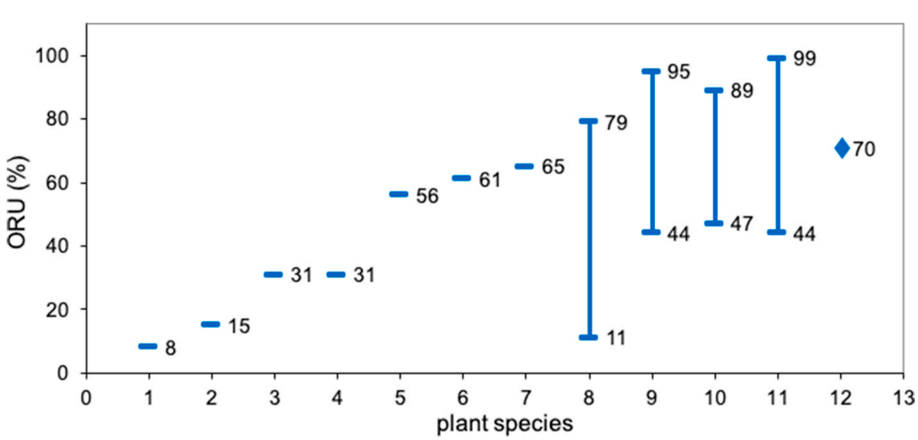

Fig. 2. Overall transfer of ^{137}Cs from hydroponic solutions to plants (%) in different experimental designs: 1 – sunflower (*Heliantus annus*) (by Soudek et al., 2004); 2 – reed (*Phragmites australis*) (by De Medici et al., 2019); 3 – wild calla (*Calla palustris*) (by Rinaldi et al., 2017); 4 – poplar (*Populus simonii*) (by Soudek et al., 2004); 5 – cress (*Lepidium sativum*) (by Marčiulioniene et al., 2008); 6 – vetiver (*Chrysopogon zizanioides*) (by Singh et al., 2008); 7 – amaranth (*Amaranthus chlorostachys*) (by Borghei and Arjmandi, 2014); 8 – Siam weed (*Chromolaena odorata*) (by Singh et al., 2009); 9 – tobacco (*Nicotiana tabacum*) (by Guldanová et al., 2010); 10 – calendula (*Calendula alata*) (by Borghei et al., 2011); 11 – giant milky weed (*Calotropis gigantea*) (by Eapen et al., 2006); 12 – onion (*Allium cepa*) (the data of the present study).

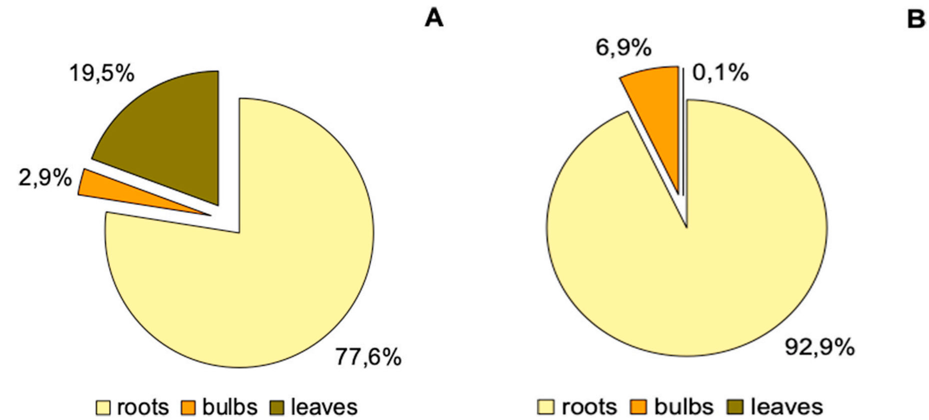

Fig. 3. Distribution of ^{137}Cs (A) and ^{243}Am (B) between organs of onion after 27 days of hydroponic cultivation.

research were fully consistent with these expectations: the $TLF^{Am-243}{}_{bulbs}$ was <0.05, and further translocation of the radionuclide from bulbs to leaves was even ≈50 times less, that emphasised negligible mobility of $^{241(243)}Am$ in xylem sap transport.

Taking into account the structure of onion biomass and corresponding values of the specific activity of radionuclides in roots, bulbs and leaves, it can be highlighted that almost 80% of ^{137}Cs and more than 90% of ^{243}Am were accumulated in roots (Fig. 3), despite the fact that the share of roots themselves in the total onion biomass corresponded to only 26%. Similar results were obtained by Bystrzejewska-Piotrowska and Urban (2004) for hydroponically cultivated onion, where only 8% of ^{137}Cs was translocated from roots and bulbs to leaves.

The so-called rhizofiltration effect seen in onion under the exposure to the radionuclides might be considered a way of adapting plants to pollution. Regarding the mechanism of rhizofiltration in relation to americium and other transuranic elements, a consensus exists amongst researchers on its bounding to solid components of the epidermal cell wall and/or chelation and deposition with the defence polymers which prevents $^{243(241)}Am$ translocation from roots into shoots (Bondareva et al., 2010; Kabata-Pendias, 2010; Serre et al., 2019). The mechanism of ^{137}Cs binding in the intra/intercellular space of plants has not been completely resolved and requires additional investigations. To gain greater insight into ^{137}Cs immobilization in plant roots Kawachi et al. (2016) used an original γ-camera that traced the absorption of ^{137}Cs from a hydroponic solution into intact soybean plants, and they noticed extremely slow radiocaesium translocation to the shoot compared to the bulk water-flux from root to shoot; so the researchers speculated a strong interaction of ^{137}Cs with the surrounding root tissues during vascular transport. *In vivo* NMR experiment revealed ^{133}Cs location in the cytoplasm and vacuolar intracellular compartments of maize root tips (Pfeffer et al., 1990), while the investigation of aerial parts of *A. thaliana* showed a possibility for Cs^+ binding in chloroplasts with simultaneous presence in cytoplasm of leaf cells (Le Lay et al., 2006). Anatomical study of *C. album* showed the immobilization of Cs^+ in the parenchyma as a crystalline structure (Borghei et al., 2011), while chelation of Cs^+ ions is supposed to be a way for the tolerance of Indian mustard (*B. juncea*) to abiotic stress (Lai and Luo, 2019).

3.3. *Visualization of ^{137}Cs and ^{243}Am incorporation into onion organs via autoradiography images*

Principal patterns of quantitative γ-spectrometric data on the ^{137}Cs and ^{243}Am distribution in onion organs were confirmed on a qualitative basis of digital autoradiographic images (Fig. 4). First, after 27 days of onion cultivation in ^{137}Cs-containing solution with specific activity of 250 kBq L^{-1} the radionuclide was manifested in all onion organs – roots, bulbs and leaves. Earlier autoradiographic images, reflecting root uptake and further translocation of ^{137}Cs into sunflower, reed and poplar grown in hydroponics, were shown by Soudek et al. (2004, 2006). Second, after cultivation of onion in ^{243}Am-containing solution with the specific activity of 9 kBq L^{-1} a contrasting imprint was obtained only for roots, whereas even the diurnal measurement of the bulbs and leaves did not allow to get distinct autoradiographic images due to very small amounts of the radionuclide incorporated in these organs. Such results were also noted previously by Soudek et al. (2011) on uranium transfer from hydroponic solutions to twenty other plant species.

When interpreting of autoradiographic images of onion bulbs and leaves, it was assumed that the contrast of the pictures was mainly depended on the thickness of tissues, so there were not discovered signs of amplified ^{137}Cs accumulation in young shoots or leaf tips, as noted by Soudek et al. (2004, 2006) in sunflower (*Heliantus annus*), reed (*Phragmites australis*) and poplar (*Populus simonii*); at least with the image resolution available in this study.

At the same time autoradiography allowed finding some additional features of the radionuclide's distribution within onion roots. Namely, after onion exposure to ^{137}Cs, darker sections, 5–7 cm in length, were visible near the root tips, which indicated an increased level of the radionuclide accumulation there. Autoradiographic visualization of the spatial distribution of ^{243}Am in onion roots was even more impressive: there were numerous fine radioactive "hot spots" localized closer to the root apex. In both cases, it seems that after penetration into the onion root tissue, the radionuclides, especially ^{243}Am, were largely immobilized *in situ*. Active bioaccumulation of the radionuclides near the tips of roots, *i.e.* in the most important functional zones of elongation and maturation, could disturb cell division in the roots, impair plant nutrition and lead to resulting changes in the morpho-physiological traits of plants.

3.4. *Biometric traits of onion grown in ^{137}Cs- and ^{243}Am-containing solutions*

Biometric – morphological and biomass – traits could be considered as indicators of the biochemical and physiological processes in plants evolving in response to radioactive/chemical stress. It is interesting to note in this connection that under exposure of onion to the radionuclides the biometric traits of the plants changed mainly under the influence of ^{243}Am (Table 3), whilst the specific activity of ^{243}Am in hydroponic solution was almost 30 times less than that of ^{137}Cs, and simultaneously TF^{Am-243} was ≈5 times lower than TF^{Cs-137}. The key reason for the different phytotoxicity of the radionuclides could be the specificity of the effects induced by γ- and α-ionizing radiation. Similarly, the hazardous effects of ^{241}Am (α-emitter) and weak – ^{133}Ba (γ-emitter) in hydroponic solutions with comparable activity concentrations 2.2 and 1.5 kBq L^{-1}, respectively, on Arabidopsis roots were previously established by Biermans et al. (2014). The authors concluded that while the photons emitted by ^{133}Ba traveled long distances and escaped a plant, α-particles

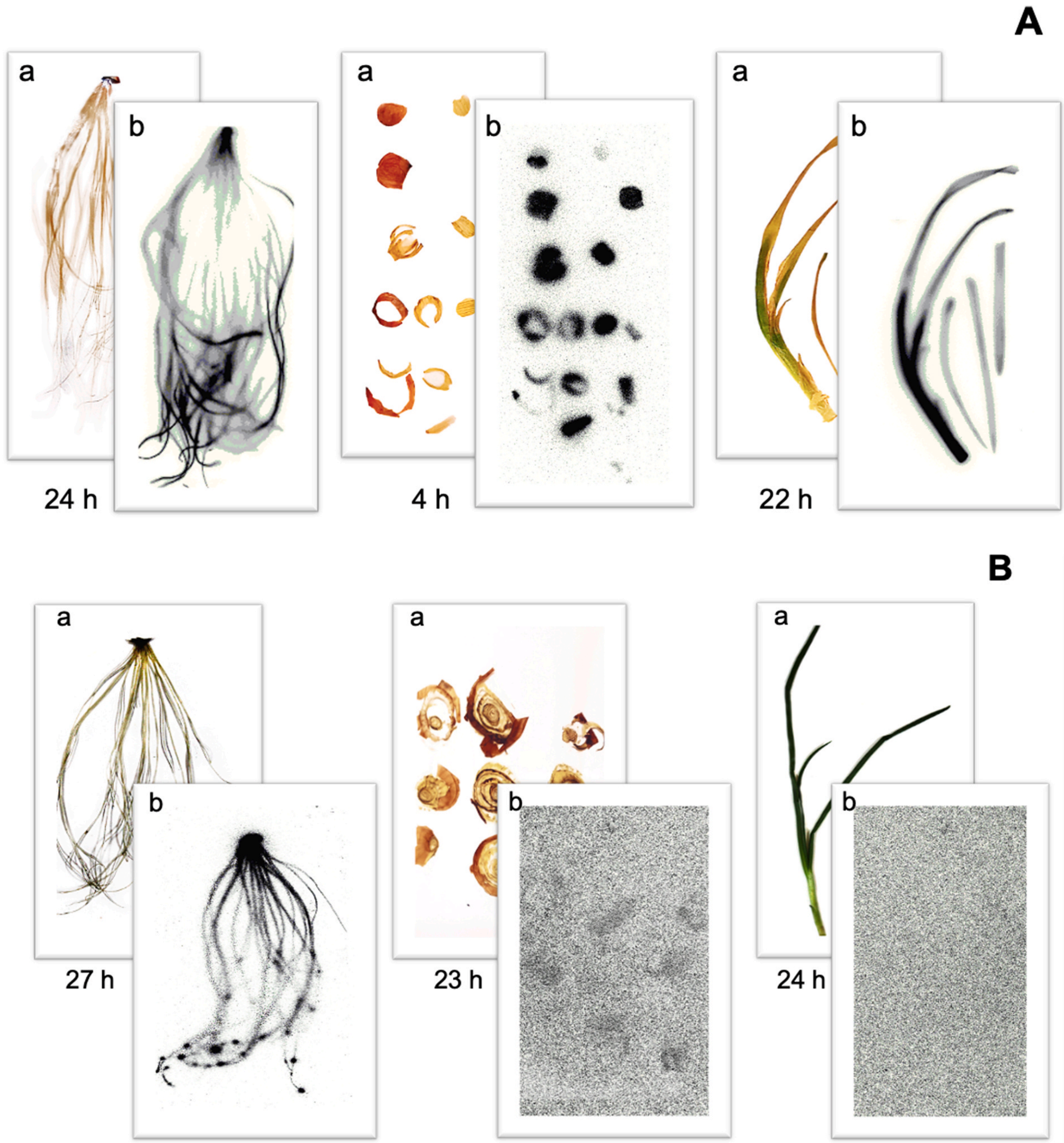

Fig. 4. Autoradiographic visualization of ^{137}Cs (A) and ^{243}Am (B) bioaccumulation in onion roots, bulbs and leaves after 27-day hydroponic cultivation: (a) photography, (b) autoradiography. Exposure time in hours is shown next to each autoradiography.

from ^{241}Am caused a large number of ionizations at a very short distance right within the root, directly adversely affected root growth, and indirectly inhibited carbon assimilation and photosynthesis.

As expected, plant roots were the main phytotoxic targets when cultivation of onion with ^{137}Cs and ^{243}Am due to a combination of external ionizing radiation from solutions and elevated level of the radionuclide's bioaccumulation in the roots themselves. When uranium was used in such an experiment, then in subtoxic concentrations it stimulated primary root growth of *A. taliana* and reduced secondary root formation, while at a toxic level it suppressed the growth of primary roots and stimulated the formation of secondary roots (Serre et al., 2019). In the present study, cultivation of onion with subtoxic ^{243}Am activity level significantly influenced the processes of lateral roots formation and, at the same time, their elongation. RN value reduced 1.7 times, and RL_{avg} as well as RL_{max} values – 1.3 times. The most sensitive indicators seemed to be RL_{sum}, which reduced by the factor 2.2, and especially RA, which depended on both RL_{sum} and RN and decreased 2.6 times. In general, under the influence of ^{243}Am, the processes of growth and development of onion roots were essentially suppressed, which led to a change in all geometric features in comparison with the control, with the exception of invariant average RD value. Thus, ^{243}Am phytotoxicity was markedly reflected on the roots formation and elongation, but not on their cross-section diameter. The last was different from the effect of some heavy metals on Arabidopsis, which consisted in thickening of the roots due to the larger number and size of cells in the epidermis and cortex (Staňová et al., 2012).

Root biomass traits of onion exposed to ^{243}Am – RFB^{Am-243}, RDB^{Am-243}, and $RDMC^{Am-243}$ – insignificantly went down by nothing more 7–11%. Taking into account a significant decrease in the total volume of the roots together with small changes in their biomass, an

Table 3
Biometric traits of onion under cultivation with ^{137}Cs- and ^{243}Am-containing hydroponic solutions (all characteristics – per one plant).

Biometric characteristics	^{137}Cs-treatment	^{243}Am-treatment	Control
Number, length and surface area of leaves			
LN	3.1 ± 0.2[a]	3.3 ± 0.5	3.0 ± 0.8
LL_{max}, cm	31.0 ± 2.6	30.3 ± 2.0	31.1 ± 4.2
LA, cm^2	58.8 ± 13.4	43.9 ± 12.8	47.3 ± 6.4
Number, length, diameter and surface area of roots			
RN	29.3 ± 3.7[ab]	18.8 ± 3.5[b]	31.2 ± 2.0[a]
RL_{avg}, cm	25.5 ± 1.5[a]	19.1 ± 3.4[b]	25.7 ± 0.4[a]
RL_{max}, cm	35.4 ± 3.0[a]	28.5 ± 1.7[b]	37.6 ± 5.1[a]
RL_{sum}, cm	746 ± 94.2[a]	363 ± 99.2[b]	803 ± 53.2[a]
RD, cm	0.11 ± 0.01	0.11 ± 0.02	0.12 ± 0.04
RA, cm^2	158 ± 28.1[a]	75.1 ± 17.9[b]	192 ± 48.9[a]
Fresh biomass			
LFB, g	2.5 ± 0.5	2.2 ± 0.4	2.3 ± 0.6
BFB, g	1.7 ± 0.5	2.1 ± 0.3	1.9 ± 0.6
RFB, g	1.7 ± 0.5	1.3 ± 0.3	1.4 ± 0.3
TFB, g	5.8 ± 1.4	5.6 ± 0.9	5.6 ± 1.1
Dry biomass			
LDB, mg	133 ± 27.7	129 ± 22.7	133 ± 35.9
BDB, mg	117 ± 29.4	129 ± 28.3	144 ± 47.5
RDB, mg	89.5 ± 23.9	84.2 ± 14.2	94.3 ± 18.8
TDB, mg	339 ± 67.0	338 ± 57.5	372 ± 78.7
Dry-matter content			
LDMC, mg g^{-1}	54.9 ± 7.1	58.4 ± 2.8	61.2 ± 2.6
BDMC, mg g^{-1}	75.3 ± 17.5	62.4 ± 6.4	81.0 ± 15.1
RDMC, mg g^{-1}	54.3 ± 2.6[a]	64.9 ± 5.8[b]	72.6 ± 7.0[b]
TDMC, mg g^{-1}	59.8 ± 6.0	61.4 ± 3.7	69.7 ± 5.5
Area/biomass and length/biomass indexes			
SLA, cm^2 mg^{-1}	0.39 ± 0.06	0.34 ± 0.02	0.31 ± 0.03
SRA, cm^2 mg^{-1}	1.63 ± 0.40	1.16 ± 0.20	1.73 ± 0.16
SRL, mm mg^{-1}	7.74 ± 1.82	5.50 ± 0.97	7.86 ± 0.55

[a] The numerical values presented show arithmetic mean ± confidence interval at $\alpha = 0.05$ at least for 6 biological replicates for processing of digital photos of onion leaves and roots and for 10 biological replicates for biomass measurements.

[b] Different letters next to the numerical values show statistically significant differences between the biometric characteristic of the control and ^{137}Cs- and ^{243}Am-treatment after Tukey's HSD multiple comparison at $\alpha < 0.05$; numerical values without accompanying letters indicate no significant differences between these groups.

increase in the density of the root tissues could be assumed. As a whole, root development and elongation parameters were found to be especially sensitive to ^{243}Am toxicity, as was often noted for phytotoxic effects of heavy metals (Bagur-González et al., 2011), while biomass traits of onion were more resistant. The integrated area/biomass and length/biomass indexes for onion roots under ^{243}Am-treatment eventually decreased by 30–33%, but the significance of these differences was not confirmed statistically.

The mechanism of the phytotoxic impact of ^{137}Cs on onion roots was completely different: the processes of root elongation were either not disturbed or only slightly suppressed, whereas the basic characteristics for calculating the dry-matter content changed with moderate opposite tendencies – an increase in RFB^{Cs-137} and a decrease in RDB^{Cs-137}. The resulting statistically significant decline in the value of $RDMC^{Cs-137}$ by 1.3 times indicated a corresponding rise in the root water content (the inverse value, calculated simply as 1000 – $RDMC^{Cs-137}$) and revealed the influence of ^{137}Cs on hydration degree in root tissues. Moreover, the preservation of the architectural volume of onion roots with a simultaneous decrease in the value of $RDMC^{Cs-137}$ could mean a possible reduction in the density of their tissues under ^{137}Cs-treatment.

Similar adverse influence on plants' water status has been revealed for Cd, the most toxic amongst heavy metals, which ultimately limited nutrient intake to plant roots (Qin et al., 2020). Besides, Bystrzejewska-Piotrowska and Urban (2003) detected severe osmotic stress in the watercress (*Lepidium sativum*) seedlings induced by 1 mM ^{133}Cs solution, however, accompanied by a drop of water uptake intensity by root and the dehydration of the whole plant. It seems like the concrete phytotoxic effect of $^{137(133)}$Cs essentially depend on the plant species and experimental design. In particular, there was a decrease in the growth of roots and leaves of lettuce (*Lactuca sativa*) at 15 mM ^{133}Cs concentration in hydroponic solution (De Medici et al., 2019), as well as a decrease in root length and biomass of crested wheatgrass (*Agropyron spicatum*) and cheatgrass (*Leymus cinereus*) under soil treatment with ^{133}Cs 50 mg kg^{-1} (Cook et al., 2009). But a number of researchers reported neither symptoms of toxicity, nor changes in biometric characteristics of the investigated test-crops (*Lactuca sativa*, *Sorghum bicolor*, *Panicum virginatum*, *Amaranthus chlorostachys*, *Chenopodium album*, *Calendula alata*) in hydroponic or soil pot experiments under $^{137(133)}$Cs exposure up to 100 kBq kg^{-1} or 45 mM L^{-1} (Entry and Watrud, 1998; Moogouei et al., 2017; Šuňovská et al., 2012; Wang et al., 2012). In a study of Burger et al. (2019a), the root length of *P. major* was not affected, but the reduction in biomass was observed over a wide range of ^{133}Cs concentrations used. It should be remarked that in the present investigation the selected level of ^{137}Cs in the hydroponic solution was two orders of magnitude lower than that which is usually considered chemically toxic for stable ^{133}Cs (≈200 μM L^{-1}); therefore, the observed effect of radiocaesium on the water regime of plants was most likely determined by the impact of γ-radiation.

There is no doubt that the revealed changes in the architecture of onion roots under the influence of the radionuclides reflected deep molecular and cellular events in plants as a whole (White et al., 2013). Nevertheless, after 27 days of onion exposure to ^{137}Cs and ^{243}Am, all geometric and biomass characteristics of bulbs and leaves did not statistically differ from the control. Only a statistically insignificant reduction of the $LDMC^{Cs-137}$ value by ~ 10% could be considered as a slight continuation of the trends towards an increase in relative hydration and a decrease in the density of leaf tissues, which were more clearly noted for onion roots. Root-to-shoot translocation of absorbed ^{243}Am, in turn, was very limited, therefore, no clear changes in the biometric characteristics of onion leaves and bulbs were detected.

At the level of the whole organism, the fresh onion biomass yield did not undergo changes under the influence of the radionuclides as compared to the control, and the average values of total onion dry biomass and total dry-matter content insignificantly decreased by 9–14% when growing plants both in ^{137}Cs- and in ^{243}Am-containing solutions.

4. Conclusions

The morphophysiological response of onion to chronic stress under cultivation in low-mineralized neutral hydroponic solutions with ^{137}Cs (250 kBq L^{-1}) or ^{243}Am (9 kBq L^{-1}) revealed similarity and difference in plant phytotoxicity and tolerance caused by these long-lived artificial radionuclides. Thus, at $TF^{Cs-137}{}_{tot}$ ≈400, effective root uptake of ^{137}Cs through non-specific ion transport channels, similar to those of essential K^+ and NH_4^+, led to the fact that ≈70% of the radionuclide was incorporated into the onion biomass during the 27-day cultivation period. Root uptake of ^{243}Am by onion, in turn, was essentially limited ($TF^{Am-243}{}_{tot}$ ≈80; and ≈14% of the total amount in "hydroponic medium – plant" system), apparently due to the lack of similarity with essential nutrients and a gradual reduction of ^{243}Am bioavailability owing to complexation with root exudates.

However, after being absorbed, both radionuclides mainly accumulated in the roots of onion (≈80% of ^{137}Cs and more than 90% of ^{243}Am), therefore, rhizofiltration strategy was realized to protect, to some extent, the edible aerial parts against radioactive contamination. Simultaneously, bioaccumulation of the radionuclides in onion roots with a relative increase in concentration near the tips, apparently, influenced the further growth and development of plants, especially the biometric traits of their roots.

Despite the lower amount of ^{243}Am transfer into onion as compared to ^{137}Cs, phytotoxic effects in roots were considerably more pronounced under such treatment: a statistically significant decrease in the number and average length of roots (by 1.3 times), total root length and surface area (by 2.2–2.6 times) were observed. An increase in the density of root tissues could be also assumed under onion exposure to ^{243}Am. The specificity of ^{137}Cs-treated onion consisted of moderate the violation in the degree of hydration of root tissues and a corresponding decrease in the dry-matter content in the roots (by 1.3 times). As a whole, biometric traits of onion roots exhibited sensitivity to ^{137}Cs and ^{243}Am chronic exposure and could serve as appropriate easy measured indicators of the phytotoxicity of the radionuclides.

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Author Contribution

Tatiana Paramonova: Conceptualization, Methodology, Formal analysis, Investigation, Writing - original draft, Visualization; Natalia Kuzmenkova: Conceptualization, Investigation, Writing - original draft, Visualization; Maria Godyaeva: Methodology, Software, Investigation, Writing - original draft, Visualization; Ekaterina Slominskaya: Methodology, Investigation, Visualization.

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.

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