

Bioaccumulation of Radionuclide Metals in Plants: A Case Study of Cesium

S. Mehdi Borghei, Reza Arjmandi and Roxana Moogouei

Abstract Phytoremediation is a new and fast-developing technology with non-destructive properties and ideal for removal of heavy metals and radionuclides from industrial effluents. Many plant species have high efficiency for adsorption and uptake of cations including heavy metals and radionuclides from contaminated soil and water. Plant screening and selection for remediation purposes is considered an important and a major step for implication of this technique. Taking into account geological characteristics and climatic conditions of a particular geographic area, most suitable plant species must be selected and implemented to achieve high rates of remediation. In this study, three plant species (*Amaranthus chlorostachys* var. *Chlorostachys*, *Calendula alata* Rech. F., Fl. Iranica, and *Chenopodium album*) were studied with the objectives to evaluate their potential for uptake of cesium from wastewater containing cesium salt. At the first step, plant seeds were selected and grew in hydroponic system using “Hoagland” solution. “Hoagland” solution is a standard medium made from distilled water enriched with specified nutrients for plant growth studies. After 2 months, plants were incubated in solutions with three different concentrations of stable cesium. The first set of experiments were carried out using four containers including a control solution containing distilled water free of Cs, and three other distilled water solutions containing 0.5, 2, and 5 mg l⁻¹ of CsCl, respectively. In the second set of experiments, standard Hoagland solution was used. *A. chlorostachys* and *C. album* plants were placed in containers filled with Hoagland medium mixed

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with equal volume of cesium solution made from dissolving CsCl in distilled water, so that the final concentration of CsCl was 1 mg l^{-1} in first container and 2.5 mg l^{-1} in the second. For each set of experiments, several similar containers were used to ensure validity of results. Experiments were arranged in randomized block design for a period of 15 days. Then, *C. album* that showed to be an effective accumulator of cesium was selected for anatomical studies. After separation, aerial organs (stem, leaf) of *C. album* plant were laid in a solution of alcohol and glycerin for fixation and anatomical investigations. The concentration of cesium in the growth solution, (Hoagland diluted with distilled water containing cesium), and in plants (dried and digested in acid) was measured by atomic absorption spectrophotometry (Varian Spectra AA-55B). The results of the present study showed that *A. chlorostachys* remediated $65 \pm 4.11 \%$ of cesium from simulated wastewater. Efficiency of *C. alata* in phytoremediation of cesium chloride (5 mg l^{-1}) and Hoagland medium was $89.35 \pm 0.25 \%$. Comparison of plants remediation potentials showed higher efficiency of *A. chlorostachys* while in bioaccumulation potential comparison, *A. chlorostachys* showed higher efficiency. Anatomical changes studies of *C. album* plants showed that important change was increase in crystals (entering cesium to crystalline structure) quantity in stem parenchyma and their color embrace, due to cesium uptake. This tolerant plant converted cesium ions to crystals molecules in shoots. Plants grew healthy in contaminated environments and the remediation efficiency of cesium reached close to 90% in *C. alata* when cesium salt concentration was 2.5 mg l^{-1} and Hoagland solution was used. It was concluded that in a proper culturing condition, all these plants are tolerant to radionuclides and could be suitable candidates for remediation of radionuclide wastes. Consequently, it was proved that phytoremediation could be a complimentary treatment technique for removal of radionuclides from waste discharges at nuclear sites.

Keywords Phytoremediation • Cesium • Plant species • Solution

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1 Introduction

The development of nuclear energy power plants and the use of radio nuclides for many industrial, medical, and also research projects could cause major radionuclide contamination of the environment and may result in health problems for several decades if no special remediation is undertaken (Vandenhove 2013). Although various technologies for remediation of contaminated soil, sludge, sediment, surface water, groundwater, and wastewater have been used, they are usually very expensive and have secondary impacts on the environment. In case of water and wastewater, the efficiency of remediation and cost-effectiveness are major considerations to strategy formulations (Salt et al. 1998; Eapen et al. 2006; Zhang et al. 2007; Chakraborty et al. 2007; Singh et al. 2008). Phytoremediation is a new and fast-developing technology with nondestructive properties and ideal for managing this type of effluents. Many plant species have been identified as hyperaccumulators; that is, they have the ability to accumulate high concentrations of metals, without impact on their growth and development (Xiong 1997). Many studies have examined the ability of plants to remediate a variety of elements from diverse media. The achievement of phytoremediation depends on plant growth rate and obtaining high metal concentrations in plant shoots (Alloway et al. 1990; Tanhan et al. 2007). Plants uptake system is defined as a system that involves ions uptake from environment. The main components of the species-specific uptake system are transporters and channels (Maestri et al. 2010). Depending on the plants uptake rate and distribution of elements within the plant organs, their content and distribution is significantly diversified (Verkleij and Schat 1990). Moreover, plant concentrations of metals may be influenced by a variety of conditions. Not only pH but also other ions concentrations and environmental conditions may interact with uptake of elements and sometimes change the growth rate of plants (Massas et al. 2010). In recent studies, there seems to be an enduring interest in selecting native plants that are tolerant to pollutants and many researchers have evaluated the phytoremediation potential of native plants under field conditions (McGrath and Zhao 2003). Experimental real-life studies are necessary and may have to include a range of contaminant concentrations, mixtures of various contaminants, and different experimental treatments to prove the sustainability of this technique for efficient remediation. Plant selection is based on growth rate, contaminant translocation, accumulation potential, and tolerance to contaminants. Singh et al. (2009) have found that plants belonging to Chenopodiaceae, Amaranthaceae, and Asteraceae families are effective remediators of many cations including heavy metals. The main purpose of this work has been largely to evaluate the potential of

Chenopodium album, *Amaranthus chlorostachys*, and *Calendula alata* to phyto-remediate stable cesium as an analog of radioactive Cs. Moreover, metal concentration in shoots of plants was compared with those in roots. *C. album*, *A. chlorostachys*, and *C. alata* are fast-growing plants that grow on wide geographic allocations in arid and semiarid regions of the world. Many of plant species have a high efficiency in radionuclide uptake from polluted environment (McCutcheon and Schnoor 2003; Dabbagh et al. 2008). Radionuclide's bioaccumulation potential is different in various plants. Some plants that have been used for this purpose are shown in Table 1. Plant screening and selection for remediation purposes is the most important step of this method. Based on geographical and climatic characteristic of each area, plant species can be selected and used.

Radionuclides occur naturally in soil. A radionuclide is described as an atom with an unstable nucleus. When radionuclides decay, they produce ionizing radiation. Radioactive substances decay with a characteristic "half-life," which is described as the time required for reducing the activity of a radioactive particle to half of its initial activity. Those resources that produce radiation over long periods have adverse effects on human health. The main categories of ionizing radiation emitted as a consequence of radioactive decay are alpha, beta, gamma, radiation, and fission with emission of fragments and neutrons. Radioactive decay can neither be inhibited nor accelerated. Since half-lives can be many thousands or millions of years, radionuclides must be safely stored meanwhile. While radionuclides appear physically in the environment, those categorized as injurious are usually of anthropogenic sources, or enhanced by human activities and dispersed through industrial applications. The Nuclear Regulatory Commission classified radioactive materials into two categories: high-level and low-level classes. High-level radioactive substances mainly originates from the fuel used by a reactor to produce electricity, while low-level wastes include material that either has a short decay time or has become contaminated with or activated by nuclear materials. Uranium mining and nuclear reactors are common sources of radionuclide production. The processing of uranium in nuclear reactors similarly produces isotopes such as cesium 137 and strontium 90, which take a considerable long time to decay. In addition, uranium 238 decays to form radium 226 that has a half-life of 1,600 years. A lot of these radioisotopes stay active in industrial by-products and wastes for long periods of time. Other usually encountered radionuclides include cobalt 60, plutonium, radium, radon, technetium 99, thorium, and uranium. Radionuclides are used for a variety of purposes that can be highly beneficial or equally high health hazards if disposed and discharged inefficiently. Radionuclides are useful for their chemical properties and are used in biomedicine in the diagnosis, treatment, and research into diseases. Radionuclides that release gamma rays can be used as tracers to monitor bodily states and the functioning of organs, while radium and radon can be used in the treatment of cancerous tumors. Furthermore, in scientific research into genetics, radionuclides allow researchers to label molecules and study processes such as DNA replication, and transformation.

Table 1 Candidate plants used for phytoremediation of radionuclides

Plant (scientific name) or family or genus	English name
Cs	
<i>Acer rubrum</i> , <i>Acer pseudoplatanus</i>	Red maple
<i>Agrostis spp.</i>	Bent grass
<i>Amaranthus retroflexus</i>	Red-root Amaranthus
<i>Amaranthus chlorostachys</i>	Red-root Amaranthus
<i>Brassicaceae</i>	Mustards, mustard flowers, crucifers, or cabbage family
<i>Brassica juncea</i>	Indian mustard
<i>Cerastium fontanum</i>	Big chickweed
<i>Beta vulgaris</i> , <i>Chenopodiaceae</i>	Beet, Quinoa, Russian thistle
<i>Cocosnucifera</i>	Coconut palm
<i>Eichhornia crassipes</i>	Water hyacinth
<i>Eragrostis bahiensis (Eragrostis)</i>	Bahia love grass
<i>Eucalyptus tereticornis</i>	Forest red gum
<i>Festuca arundinacea</i>	Tall fescue
<i>Festuca rubra</i>	Fescue
<i>Helianthus annuus</i>	Sunflower
<i>Larix deciduas</i>	Larch
<i>Liquidambar styraciflua</i>	American sweet gum
<i>Liriodendron tulipifera</i>	Tulip tree
<i>Lolium multiflorum</i>	Italian ryegrass
<i>Lolium perenne</i>	Perennial ryegrass
<i>Panicum virgatum</i>	Switch grass
<i>Phaseolus acutifolius</i>	Tepary beans
<i>Phalarisarundinacea L.</i>	Reed canary grass
<i>Picea abies</i>	Spruce
<i>Pinus radiata</i> , <i>Pinus ponderosa</i>	Monterey pine, Ponderosa pine
<i>Sorghum halepense</i>	Johnson grass
<i>Trifolium repens</i>	White clover
<i>Zea mays</i>	Corn
<i>Sorghum</i>	Sorghum
<i>Fagopyrum esculentum</i>	Buckwheat
<i>Chromolaena odorata</i>	Siam weed
<i>Beta Vulgaris</i>	Beet
<i>Salsola kali</i>	Prickly saltwort
<i>Calotropis gigantea</i>	Sodom apple
<i>Phlox hoodii</i>	Spiny phlox
<i>Purshia tridentate</i>	Antelope brush
<i>Artemisia vulgaris</i>	Mugwort
<i>Eriogonum umbellatum</i>	Polygonaceae
<i>Lomatium foeniculaceum</i>	Sulfur flower buckwheat

(continued)

Table 1 (continued)

Plant (scientific name) or family or genus	English name
<i>Castilleja angustifolia</i>	Northwestern Indian paintbrush and desert Indian paintbrush
<i>Taraxacum officinale</i>	Common dandelion
<i>Eichhornia crassipes</i>	Water hyacinth
<i>Cordylanthus ramosus</i>	Bushy bird's beak
Co	
<i>Haumaniastrum robertii</i>	Copper flower
<i>Thlaspicae rulescens</i>	Alpine pennycress
<i>Acer rubrum</i>	Red maple
<i>Thlaspicae rulescens</i>	Alpine pennycress
Pu	
<i>Liquidambar styraciflua</i>	American sweet gum
<i>Liriodendron tulipifera</i>	Tulip tree
Ra	
<i>Poaceae various species</i>	
<i>Dryopteris scottii</i>	
<i>Asteraceae various species</i>	
Sr	
<i>Acer rubrum</i>	Red maple
<i>Brassicaceae</i>	Mustards, mustard flowers, crucifers or, cabbage family
<i>Beta vulgaris, Salsola Kali</i>	Beet, Quinoa, Russian thistle
<i>Eichhornia crassipes</i>	Water hyacinth
<i>Eucalyptus tereticornis</i>	Forest redgum
<i>Helianthus annuus</i>	Sunflower
<i>Liquidambar styraciflua</i>	American sweet gum
<i>Liriodendron tulipifera</i>	Tulip tree
<i>Lolium ultiflorum</i>	Italian ryegrass
<i>Pinus radiata, Pinus ponderosa</i>	Monterey pine, Ponderosa pine
<i>Apiaceae (a.k.a.Umbelliferae)</i>	Carrot or parsley family
<i>Fabaceae (a.k.a.Leguminosae)</i>	Legume, pea, or bean family
<i>Vetiveria zizanoides</i>	Vetiver, Khas-Khas
U	
<i>Amaranthus</i>	
<i>Brassica juncea</i>	Brown mustard
<i>Brassica chinensis</i>	Bok choy
<i>Brassica narinosa</i>	Tatsoi
<i>Eichhornia crassipes</i>	Water Hyacinth
<i>Helianthus annuus</i>	Sunflower
<i>Juniperus</i>	Juniper
<i>Picea mariana</i>	Black spruce
<i>Quercus</i>	Oak
<i>Salsola kali</i>	Russian thistle (tumble weed)

(continued)

Table 1 (continued)

Plant (scientific name) or family or genus	English name
<i>Salix viminalis</i>	Common Osier
<i>Silene vulgaris</i> (a.k.a. <i>Silene cucubalus</i>)	Bladder campion
<i>Zea mays</i>	Maize
<i>Helianthus annuus</i>	Sunflower
<i>Brassica juncea</i>	Mustard greens, Indian mustard, Chinese mustard, or Leaf mustard
Ti	
<i>Phlox hoodi</i> <i>Plemoniaceae</i>	Spiny phlox or carpet phlox
<i>Eriogonum polygonaceae</i>	California buckwheat
<i>Cordylanthus capitatus</i>	Clustered bird's beak
<i>Antennaria</i>	Cats foot
<i>Chaenactis</i> , <i>Asteraceae</i>	
<i>Fabaceae</i>	
<i>Microsteris gracilis</i> <i>Polemoniaceae</i>	Slender phlox

Another major usage of radionuclides, specifically the element uranium, is for energy production. Interest in Cs distribution in plants and the movement of this element in ecosystems extends back to the 1950s by the development of nuclear technologies used for energy production (Cook et al. 2007). Release of cesium nuclides in environment may be very similar to other more hazardous radionuclides present in wastes from nuclear wastes. The radioisotopes of cesium (^{134}Cs and ^{137}Cs) may be of special concern because of their similar behavior to the necessary element “K” in plants, their solubility in aquatic ecosystems, the volatilization, release and dispersal in major reactor accidents, and the great quantity and persistence of ^{137}Cs in spent fuel and reprocessed wastes (Pipíska et al. 2004; Pinder III et al. 2006). According to studies carried out by Tsukada et al. (2002) and Vinichuk et al. (2010), strong correlations exist between distribution of ^{137}Cs and stable Cs in plants. Moreover, Soudek et al. (2006) did not find any differences between the uptake of radioactive and stable Cs isotopes by *Helianthus annuus* L. Stable Cs is phytotoxic in solutions exceeding 200 mM (Borghei et al. 2011).

2 Methodology

Three plant species (*A. Chlorostachys*, *C. alata* Rech. F., Fl. Iranica, and *C. album*) were used to evaluate their potential for phytoremediation of Cs solutions and their tolerance to cesium uptake. Healthy seeds of *A. chlorostachys*, *C. alata*, and *C. album* were surface sterilized by 1 % sodium hypochlorite for 20 min. *C. alata* seeds were sown in a substrate containing perlite and vermiculite 3:1 (v/v) moistened with distilled water for 4 weeks until seedlings with two leaf pairs were

established. *A. chlorostachys* and *C. album* seeds were germinated in sand. Then, one-month-old plantlets were transplanted in plastic trays containing 10 l nutrient solutions. The composition of macro-elements per 100 l solution was as follows: 100 ml $\text{NH}_4\text{H}_2\text{PO}_4$ (115 g l^{-1}); 600 ml KNO_3 (107 g l^{-1}); 400 ml $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ (236 g l^{-1}); 200 ml $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ (246 g l^{-1}); 150 ml Fe-EDTA (5 g l^{-1}). The composition of microelements (100 ml of solution all together) also was: H_3BO_3 (0.38 g l^{-1}); $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ (0.22 g l^{-1}); $\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$ (1.02 g l^{-1}); $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ (0.08 g l^{-1}); $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$ (0.02 g l^{-1}). Solution pH was adjusted to 5.5 through 5.8 with 0.1 M NaOH or 0.1 M HNO_3 and continuously aerated with an air pump for proper mixing and oxygenation of the solution. Nutrient solutions renewed at every tenth day. Level of the solutions in trays was made up with nutrient solution when required. Each tray contained 24 plants. Plants were grown outdoors with temperature ranging from $31 \text{ }^\circ\text{C}$ through $40 \text{ }^\circ\text{C}$ (maximum daily temperature) and $17 \text{ }^\circ\text{C}$ through $28 \text{ }^\circ\text{C}$ (minimum daily temperature) with natural light during the experiment. After 4 weeks, plantlets with uniform size were selected and transferred to 1-l flasks.

2.1 Experiments Using Hydroponically Grown Plants

2.1.1 Remediation of Cs Solutions Contaminated with Cs

The roots of cultured plants were washed thoroughly with distilled water, and plants were incubated with roots immersed in 1 l solution with three different Cs concentrations. The treatment samples included: (1) control sample free of Cs, samples 2, 3, and 4 containing 0.5, 2, and 5 mg l^{-1} of CsCl, respectively. Consequently, the concentration of Cs ions in the solutions was 0.47, 1.58, and 3.95 mg l^{-1} . The experiment was arranged with each treatment in triplicate samples. The treatment group was exposed to CsCl solution for a period of 15 days in 1,500-ml flasks. pH of the solution was adjusted to 5.5. The average root lengths were 300 mm (*C. alata*), 350 mm (*A. chlorostachys*), and 200 mm (*C. album*). Those for plants shoots were 350 mm (*C. alata*), 400 mm (*A. chlorostachys*), and 300 mm (*C. album*). *C. alata* and *A. chlorostachys* plants had a massive root system. Each flask contained three plants, which represented one replicate. Plants grown in water served as control samples. Distilled water was used for solution preparation and for makeup of lost water. After treatment period, samples of solutions were drawn out from the solutions and analyzed for Cs concentrations. In all experiments, Cs contents of solutions were determined using atomic absorption spectrophotometry (Varian Spectra AA-55B).

2.2 The Percentage Metal Uptake

The percentage metal uptake was calculated.

$$\% \text{ uptake} = [(C_0 - C_1)/C_0] \times 100$$

where C_0 and C_1 are initial and remaining concentrations of metal, respectively, in solution (mg l^{-1}) (Abdel-Halim et al. 2003; Tanhan et al. 2007).

2.3 Distribution of Cs in *Calendula alata*, *Amaranthus chlorostachys*, and *Chenopodium album*

At the end of the experiment, plants were thoroughly washed with distilled water, separated into root and shoot and dried in an oven at 60 °C for 48 h. The dried samples were digested in $\text{HNO}_3\text{:HClO}_4$ (5:1, V/V) and analyzed for Cs by flame atomic absorption spectrophotometry. The concentrations of elements in the samples are reported on a dry matter basis.

2.4 Concentration Ratio

The concentration ratio (CR), defined as the ratio of metal concentrations in plant shoots to those in the roots (Gonzaga et al. 2006; Bidar et al. 2007), was calculated to check the effectiveness of plants in translocating metals to their aerial parts (Dahmani-Muller et al. 2000; Zabudowska et al. 2009).

2.5 Statistical Analysis

The experiments were performed in triplicate, and the statistical analysis was performed using statistical analysis system (SAS) software package. To confirm the variability of results, all the data were subjected to analysis of variance to consider the significance differences. More over, mean comparison between data was obtained using Duncan's test.

2.6 Anatomical Studies

After separation, aerial organs (stem, leaf) of *C. album* plant were laid in alcohol and glycerin for fixation. After a period of seven days, a handy section was

prepared and colored with multiple coloration (Carmen Zuji and methylene blue with ratio 1:10). Then, samples were photographed and investigated by Nikon microscope.

2.7 Result and Discussion

Methods for cleaning up radioactive contaminated environments are urgently needed (Vinichuk et al. 2013). Phytoremediation technique is based on the capability of various plants to remove different hazardous contaminants present in the environment.

2.8 Cs Remediation from Different Solution Using Hydroponically Grown Solution

As a result of this study, it was found that the selected plants were efficient in remediation of Cs from Cs wastes (Table 2) with different rates.

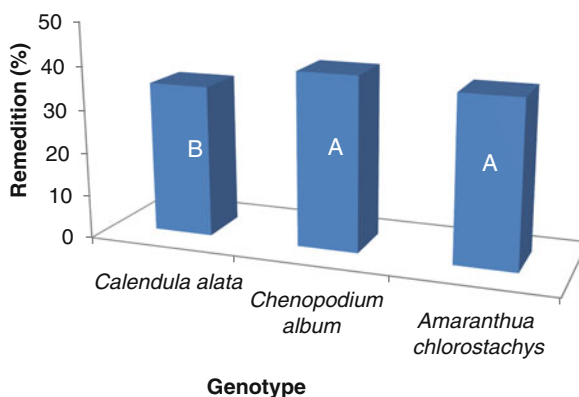
In the other experiments, when *A. chlorostachys* and *C. alata* plants were exposed to 0.5 mg l^{-1} CsCl solution, the remediation percent was calculated equal to 45 ± 8.59 and 46 ± 2.12 , respectively. Moreover, when *C. album* plants were incubated in CsCl solution in three different concentrations of 0.5, 2, and 5 mg l^{-1} , the remediation percent was 68 ± 2.12 , 39 ± 3.48 , and 52 ± 5.57 , respectively. Mean comparison analysis in remediation potential of these three plant genotypes was shown in Fig. 1. As shown in Fig. 1, *C. album* and *A. chlorostachys* had a similar behavior in cesium uptake. As presented in this study, weeds potential for phytoremediation was more than ornamental plants. Weeds were tolerant to Cs wastes. Moreover, *C. album* plants showed high potential of remediation of CsCl solution; it was selected for anatomical studies.

2.9 Cs Concentration Ratio

Cs CR in different plants was calculated in Table 3. Plants potential for phytoremediation can be assessed by CR index. In phytoextraction, radionuclide ions transfer from roots to shoots. So, plants shoots can be harvested. Tolerant plants limit movement of metals from the environment to shoots and from shoots to roots. Consequently, bioaccumulation in these plants is low. Hyperaccumulators actively uptake metals ions and transport them to their shoots. In the present study, most of the plants due to CR more than 1 are proper for cesium phytoremediation purposes. In this study, *C. alata* plants showed highest CR equal to 4.98 ± 0.04 as a

Table 2 Plant remediation potential of Cs wastes

Cs	<i>Amaranthus chlorostachys</i>	<i>Amaranthus chlorostachys</i>	<i>Calendula alata</i>	<i>Calendula alata</i>
Concentration in initial cesium solution (mg l ⁻¹)	1.58	3.95	1.58	3.95
Concentration in cesium solution after remediation period (mg l ⁻¹)	54 ± 0.06	2.30 ± 0.15	0.92 ± 0.02	1.89 ± 0.04
Remediation (%)	65 ± 4.11	41 ± 3.92	41 ± 1.59	52 ± 1.02
Concentration in initial cesium and Hoagland solution (mg l ⁻¹)	7.9	19.75	7.9	19.75
Concentration in cesium and Hoagland solution after remediation period (mg l ⁻¹)	0.90 ± 0.01	3.01 ± 0.03	1.66 ± 0.02	2.10 ± 0.05
Remediation (%)	88.56 ± 0.19	84.72 ± 0.15	78.98 ± 0.25	89.35 ± 0.25

Fig. 1 Mean comparison analysis in remediation potential of different plant genotypes

result could be used for remediation of radionuclide wastes. Concentration ratio of *C. album* during phytoremediation of Cs waste was 3.33 ± 0.03 that was more than 1 and could be account as suitable candidate. Sandeep and Manjaiah (2008) found ¹³⁴Cs transfer factor to straw and grain decreased significantly with increase in K application levels. The ¹³⁴Cs uptake was highest in spinach followed by mustard, gram, and wheat crops. The weighted transfer factor values (straw plus grain) to spinach, mustard, and gram were experimented to be 5.54, 4.38, and 2.20 times higher as compared to wheat crop.

2.10 Anatomical Changes

Anatomical structure of aerial organs during phytoremediation process and accumulation of cesium was changed. These changes increased when cesium

Table 3 Concentration ratio (CR) of cesium through plants in different cesium chloride solutions

Cesium (mg kg^{-1})	Concentration ratio (CR)							
	<i>Amaranthus chlorostachys</i>		<i>Chenopodium album</i>		<i>Calendula alata</i>			
0.47	1.58	3.95	0.47	1.58	3.95	0.47	1.58	3.95
1.76 \pm 0.01	1.07 \pm 0.03	2.19 \pm 0.05	1.06 \pm 0.05	1.19 \pm 0.02	3.33 \pm 0.03	0.47 \pm 0.04	2.84 \pm 0.06	4.89 \pm 0.04

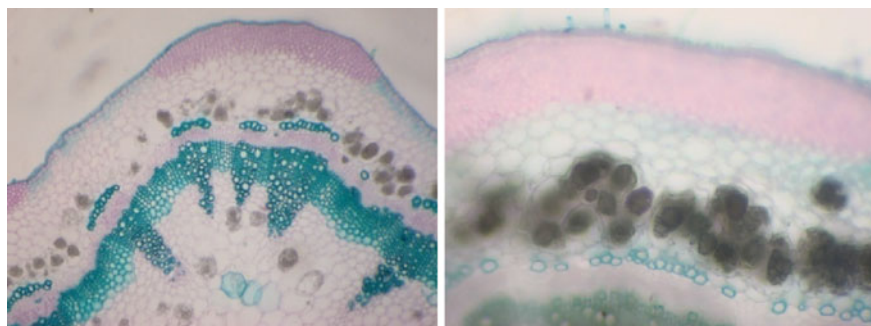


Fig. 2 Increase of crystals quantity in stem parenchyma and their color embrace after remediation of 5 g l^{-1} cesium chloride

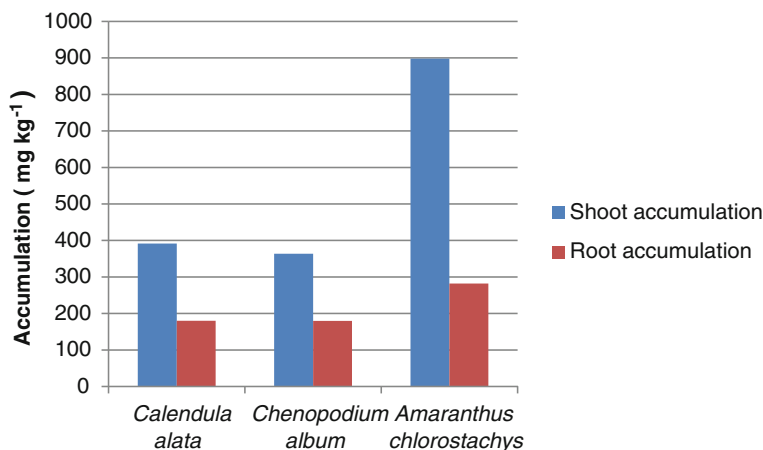
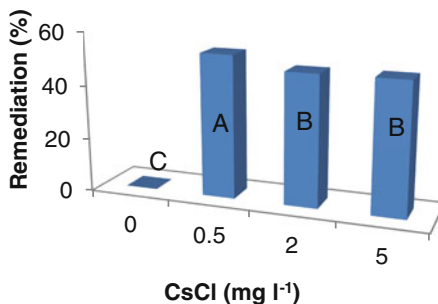


Fig. 3 Mean comparison analysis of accumulation potential between different plant genotypes

concentration in waste water intensified. As shown in Fig. 2, the most important change was increase of crystals quantity in stem parenchyma and their color embrace. The number and variation of the plant species and communities in polluted sites seemed most closely related to the activities of radioactive material and the pH value of the environment (Hu et al. 2014). In a study carried by Hu et al. 2014, the plant species in the sampling sites with moderately low activities of ²²⁶Ra and relatively high pH value showed a comparatively stable vegetation community. In our previous study, Cs accumulation in plant was studied (Moogouei et al. 2011), and further to past results, the plant accumulation potential was compared. As shown in Fig. 3, different plant genotype had significant different in phytoremediation. So, plant screening studies and experiments are important steps of using this technology. *A. chlorostachys* plants showed higher potential for cesium accumulation compared with *C. alata* and *C. album*. As shown in Fig. 4,

Fig. 4 Mean comparison analysis of Cs remediation potentials of plants in 4 different Cs concentrations



phytoremediation techniques are more efficient in lower concentration of radionuclides such as Cs. As shown in Fig. 5, more accumulation occurred during the absence of more Cs ions in the environment. Cs has unknown vital biological role for plant (Saleh 2012) while due to similarity between chemical characteristics of cesium and potassium, plants uptake both of them through one mechanism. Many transporting proteins (low affinity, inward-rectifying K channel, nonspecific, voltage insensitive cation channel, high affinity $K^+ - H^+$ symporter, voltage-dependent Ca^{2+} channels, and outward-rectifying cation channels) make possible penetration of Cs^+ across the cell membranes in plants (Bystrzejewska-Piotrowska and Bazala 2008). Furthermore, pH and temperature were important factors controlling the phytoremediation efficiency. Saleh (2012) found that the uptake rate of radiocesium from the simulated waste solution by *Eichhornia crassipes* plant is inversely relative to the initial activity content and directly relative to the increase in mass of *Eichhornia crassipes* plant and sunlight exposure, while in the present study, increase in Cs ions in solution increased Cs uptake and accumulation in plants. Moreover, as explained in Saleh (2012), research uptake rate and translocation factor were more in plants with higher mass. The uptake efficiency of ^{137}Cs present with ^{60}Co (cobalt is one of the essential trace elements necessary for plant) in mixed solution was higher than if it was incubated separately. In our study also, the presence of macro- and micronutrient ions in solution significantly enhanced remediation efficiency. Based on Saleh (2012) studies, sunlight was the most required factor for the plant vitality, growth, and radiation resistance. In the study carried by Singh et al. (2008), Vetiver grass (*Vetiveria zizanoides*) L. Nash plantlets when experienced for their potential to remove ^{90}Sr and ^{137}Cs (5×10^3 kBq l⁻¹) from solutions spiked with individual radionuclide showed that 94 % of ^{90}Sr and 61 % of ^{137}Cs could be removed from solutions after a period of 7 days. In this study and in case of ^{137}Cs , accumulation occurred more in roots than shoots. In contrary, in our study, in all three plant species, accumulation was more in shoot than root with a significant translocation factor. When experiments were performed to study the effect of analogous elements, K^+ ions reduced the uptake of ^{137}Cs , while ^{90}Sr accumulation was found to decrease in the presence of Ca^{2+} ions in *V. zizanoides* plants. In contrary, in *A. chlorostachys* and *C. alata* plants tested in this research, the presence of K^+ increased uptake of Cs from solution. Plants of

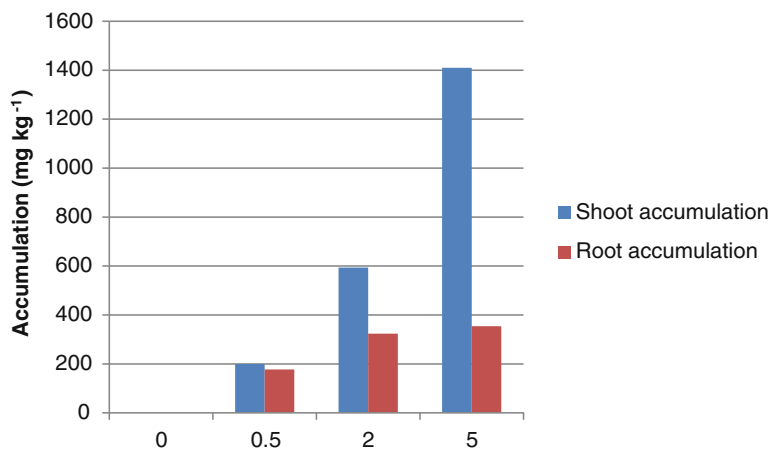


Fig. 5 Mean comparison analysis of accumulation potential through plants in 4 different Cs concentrations

V. zizanioides could also efficiently remediate radioactive elements from low-level nuclear waste, and the level of radioactivity was reduced below detection limit at the end of 15 days of exposure. As discussed by Singh (2008), *V. zizanioides* may be a potential candidate plant for phytoremediation of ⁹⁰Sr and ¹³⁷Cs. We can conclude that higher remediation potential could be achieved when using *C. alata* and *A. chlorostachys* for radionuclide phytoremediation purposes. Wang et al. (2012) had found that the ⁸⁸Sr and ¹³³Cs transfer factor values were 1.16–1.72 and 0.24–0.60, respectively, in *Raphanus sativus* L. plants. In that study, the highest ⁸⁸Sr accumulation was 239.18 $\mu\text{g g}^{-1}$ dw, while, the highest ¹³³Cs accumulation was 151.74 $\mu\text{g g}^{-1}$ dw in soil polluted with 40 mg kg^{-1} ⁸⁸Sr. The lowest ⁸⁸Sr accumulation was 131.03 $\mu\text{g g}^{-1}$ dw and the lowest ¹³³Cs accumulation was 12.85 $\mu\text{g g}^{-1}$ dw when the concentration of ⁸⁸Sr in the soil was 5 mg kg^{-1} . As there was minute influence of high concentration of ⁸⁸Sr on the total biomass of plants, therefore, the radish is one of the ideal phytoremediation candidate plants for Sr-polluted soils. In our study also, tested concentrations of Cs ions have no significant influence in total biomass of plants. These plants also showed higher CR. Li et al. (2013) have found that epiphytic *Tillandsia* plants are efficient air pollution biomonitors and usually used to monitor atmospheric heavy metal pollution, but rarely nuclides monitoring. So, they evaluated the potential of *Tillandsia usneoides* for monitoring ¹³³Cs and investigated whether Cs was trapped by the plant external surface structures. The results obtained from this study showed that *T. usneoides* was tolerant to high Cs stress. As presented in our study, they discussed that with the enhancement of Cs solution concentration, the total of Cs in plants increased significantly, which suggests that the plants could accumulate Cs quickly and effectively. Consequently, *T. usneoides* has significant potential for monitoring Cs-polluted environments. Our study showed that weeds such as *C. album* and

Fig. 6 *Amaranthus chlorostachys* plants grown in Hoagland and CsCl solution



A. chlorostachys and *A. retroflexus* were considerable potential for monitoring Cs-polluted environments. In addition, when radionuclides have been accumulated by organisms, their behavior usually reflects their similarities to macro- and micronutrients as well as nonessential elements (Markich and Twining 2012). Metabolic mechanisms moving toward homeostasis naturally make internal organism chemistry less dynamic than that in the external water columns. Many important biotic factors recognized to influence radionuclide bioaccumulation are age, gender, and size. Moreover, there are differences within and between species that reflect the natural variability within any system. These findings led to water ecosystems conservation. Scanning electron microscopy and energy dispersive spectrometer analysis in *Tillandsia usneoides* plants showed that Cs was seen in each type of cells in foliar trichomes, and the ratio of Cs in the internal disk cell was higher than that in ring cell and wing cell, which showed that the mechanism of adsorption Cs in *Tillandsia usneoides* has an active factor (Li et al. 2013). Available observation due to absorption and accumulation of Cs in *C. album* in our study classified to five groups includes:

1. Increase of crystals quantity in stem parenchyma and their color embrace.
2. Density of xylem vein groups and reduce of their dimension in stem.
3. Increasing color embrace property of xylem component in stem.
4. Decreasing cutaneous parenchyma in plants under Cs uptake stress.
5. Increasing crystalline density and colored embrace in leaf's parenchyma.

Cooperating Cs in crystal's structure and its accumulation in vacuoles of *C. album* caused plant to absorb more content of Cs radioisotopes from the environment without any serious incurred damage to plant and phytotoxicity effects. Ionic channels are activated ways for transmitting charged ion through cell membrane. Most of these channels are permeable toward some other cations such as sodium, lithium, rubidium, and Cs. Sizes of these cations are between 0.13 to 0.19 nm, which is very similar to potassium's dimension (0.27 nm), and this fact causes their spread through these channels (Tajadod and Moogouei 2012). Investigation on adsorption of Cs in this plant can be carried for air monitoring and

phytoremediation of radionuclides. REML modeling of wide-ranging CR/concentration datasets showed that the concentrations in plants of calcium, magnesium, and strontium were notably influenced by phylogeny (Willey 2013). The chemical form of a radionuclide or stable element is commonly of greater biological importance than the total concentration of it in the environment (Markich and Twining 2012). This concept was now being integrated into mechanistic frameworks such as biotic ligand and bioaccumulation models by regulators for protecting freshwater ecosystems.

3 Conclusion

Due to very high health risks, all nuclear sites have efficient systems to control their pollution and avoid environmental contamination. However, as a result of inefficiencies in treatment facilities or accidental discharges, spillage of radioactive effluents is always a cause of concern. To avoid disastrous consequences, further steps must be taken to ensure safety and upgrade preventive measures. For this goal, this study was undertaken to examine the possibility of phytoremediation as a complimentary treatment method for waste discharges from nuclear sites. Phytoremediation can be a good option for areas with low contaminations and represents a suitable and sustainable method of remediation (Willscher et al. 2013). Current research indicated that roots and shoots of *A. chlorostachys* had the most efficiency in bioaccumulation. Moreover, shoot Cs concentration was more than roots resulted in high CR of Cs. As shown in Fig. 6, plants grew healthy in contaminated area. The remediation efficiency of radionuclides reached close to 90 % in *Calendula alata* in 5 mg l⁻¹ when Cs salt was introduced into Hoagland solution. So, in a proper culturing condition, all these plants are tolerant to radionuclides and could be proper candidates for remediation of radionuclide wastes.

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