original article

Ability of phytoremediation for absorption of strontium and cesium from soils using *Cannabis sativa*

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ABSTRACT

Aims: This study is aimed at assessing the effectiveness of *Cannabis sativa* in the absorption of cesium and strontium elements from the soil.
Materials and Methods: This study was conducted in 2011, in Tehran, Iran. We employed the phytoremediation technology to refine the contamination of soil with radioactive material such as cesium and strontium. *Cannabis sativa* was selected because of its capability for potential radioactive absorption. It was planted in various soils with different concentrations of cesium and strontium (20 ppm, 40 ppm, 60 ppm, and 80 ppm), and after sufficient growth for about six months, it was separated into root, stem, and leaves for measuring the absorption of these elements in the main parts of the plant. The samples were measured by using the Inductively Coupled Plasma (ICP) method.
Results: Strontium absorption and the main parts of the plant showed a significant relationship. The percentage of strontium absorption was 45% in the root, 40% in the stem, and the minimum absorption was found in the leaves (15%), but the corresponding figure was not significant for the cesium element. A strontium concentration of 60 ppm was possibly the maximum absorption concentration by *Cannabis*.
Conclusion: Our findings suggest that strontium can be absorbed by *Cannabis sativa*, with the highest absorption by the roots, stems, and leaves. However, cesium does not reach the plant because of its single capacity and inactive complex formation.

Key words: *Cannabis sativa*, cesium, strontium, phytoremediation, radioactive materials

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INTRODUCTION

Radionuclides contaminate the environment through gases, liquids, and solids. Radioactive pollution could affect the soil and public health through a food chain.[1,2] Ecosystems all over the world have been contaminated with radionuclides caused by nuclear testing, nuclear reactor accidents, as well as nuclear power production. Radioisotope characteristics of nuclear fission, such as cesium-137 and Strontium-90, which have penetrated into the environment can become more concentrated as they enter the food chain, which could cause serious hazards to human health.[3]

Since 1991, investigations have been done on *in situ* cleaning of polluted sites by using 1700 plant species with maximum

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pollutant absorption. Of them, 47 plant indicator species are hyper accumulators, with a 37.4 – 78.1% radioactive particulate absorbing rate.[4] In 1996, a big company named Phytotech presented a report about using Sunflower and *Cannabis* for absorbing more than 95% of cesium and strontium pollution per month from a polluted site.[5]

Plants are known to absorb and accumulate Cs-137 and Sr-90, and removal of these radionuclides from polluted soils by plants can provide a trustworthy and economical method of remediation. Some advantages of the phytoremediation method are its usability for removing a wide range of organic and non-organic pollutants. It is a green technology, and if used properly, it can be an environmental friendly method. Moreover, because of its simple implementation and maintenance, it does not need either expensive equipment or expert employees. On the whole, the greatest benefit of phytoremediation is its cost-effectiveness in comparison to other environmental pollution removing methods.[4,5] It is well-documented that plants like cereals are the main sources of food pollution from Sr-90, from nuclear experiments.[6]

There are several methods for the elimination of metals from polluted soils, such as, soil washing, chemical methods, and biological methods that have engineering costs and inappropriate effects on biological activities.[7,8]

Removal of the contaminated surface soil (often up to 40 cm) or controlling radionuclides in the soils by mineral and chemical modifications is physically complicated and probably not cost-effective in practice. Phytoextraction of radionuclides by specific plant species from contaminated sites has rapidly motivated the interest of industrialists and academicians. This method is considered to be a promising bio-remediation method.[9]

Phytoremediation is a technology based on using plants, which is cost-effective as well as an environment friendly method,[10] and it is considered as an *in situ* modification method. On the other hand, using phytoremediation causes restoration of the preservation place from biological activity, physical structure, and chemical characteristics of the soil.[11,12] Strontium is an element naturally available in different parts of the environment, such as, rocks, soil, air, and water. The strontium compound moves easily in the environment on account of the water solubility of most of its compounds. A high concentration of strontium can be carcinogenic because it can harm the genetic materials in the cells. The physiological similarity of strontium with calcium causes its entrance into herbivorous milk, so if the animals graze on the polluted pastures their milk will be polluted. Consumption of cow milk, which is high in children, was forbidden in Ukraine after the Chernobyl disaster because of the risk of bone cancer after strontium absorption.[13]

After the Chernobyl disaster, low-polluted mechanisms, including reduction of radioactive elements, especially Cs-137 and Sr-90, by plant absorption and competitor cations in chemical fertilizers, on a large scale of agricultural soils was performed in East Central Europe. Radioactive material extraction by certain plant species from polluted sites has brought to the attention of factory dwellers and scientific communities that it is a biological and promising approach.[14]

For a long time now, *Cannabis* has been used as a fiber, seed oil, drug target, and a sedative drug. Industrial *Cannabis* and products of *Cannabis* plants are selected to produce a large amount of fiber. Taking into consideration the plant's potential for eliminating toxic elements and radio atoms from soils and solutions, it may be used as a method that can successfully be applied to remove Sr-90 and Cs-137 from the soil.[14]

This study aims to assess the effectiveness of *Cannabis sativa* in the absorption of cesium and strontium elements from the soil.

MATERIALS AND METHODS

This cross-sectional experimental study was conducted in 2011, in Tehran, Iran. In this investigation, *Cannabis* was used because of its potential to absorb radioactive materials, its high tolerance to metals, rapid growth, and high biomass production.[5]

Preparation of soils containing cesium and strontium

Standard solutions were prepared by using the Stoichiometry method. Nutritious soil was chosen for plant growth and the prepared solutions, with the appropriate amount of the different concentrations of elements, were added to the soil to prepare it. The amount of added solution was dependent on the soil texture. The pots for growing the plants (*Cannabis*) had to be of the same size. Considering the capacity of the pots, the amount of required soil was measured. A plastic cover was used on a flat surface for soil preparation, while transferring it into the pots, and while planting the *Cannabis* seeds. A suitable amount of solution for adding to the soil sample was determined after different experiments. It was determined that it would be 100 ml of solution for 800 g of soil in each pot. The spraying method was used for better distribution of the solution in the soil. Eight types of soil were obtained because of eight different solutions and one unpolluted soil sample was used as a control. Characteristics of radio nuclides that have attracted the most environmental concern are presented in Table 1.

Preparation of the pots

After preparation of the cesium and strontium elements, soil of the same amount was transferred to the pots. For fixing the concentrations of the elements in the soils, it was considered to have pots without any drainage hole. Therefore, after each irrigation losing the solution added to the soil would be impossible.

Small *Cannabis* seeds were scattered on the soil of each pot. Scattering the seeds on the soil was done keeping a suitable space between the seeds for their growth period.

Irrigation of pots

The irrigation frequency was twice a day. After 25 days had passed, almost all the seeds germinated and started to grow. After 38 days all the plants had a height of about 15 cm, and we disinterred them.

Removing plants from the soil

The presence of three main parts of the plant was necessary in this study. Therefore, separation of the roots from the soil was done cautiously so it came out from the soil completely. Thereafter, the roots were washed with distilled water so they were completely free of the soil particles, and all the main parts of the plant, including the leaves, stems, and roots were separated.

Drying the plant samples

The plants should have sufficient time to dry completely. In this project, due to the small sample size the natural drying method was used. The samples were placed between newspapers and after two days they were completely dry.

The dried samples were made into a powder form and transferred to separate dishes, to be used in preparing the final sample. Before preparing the final sample, the powder samples were weighed for recognizing the absorption rate of cesium and strontium.

Preparing samples for absorption measurement

The samples were measured by Inductively Coupled Plasma (ICP) (ICPMS; Perkin Elmer, Uberlingen, Germany). The samples had to be in solution form, therefore, we used 1g of each sample dissolved in 5 ml of pure nitric acid and brought it to a volume of 15 ml with distilled water. The prepared samples, with a volume of 15 ml, were put in the machine. Then strontium and cesium samples were measured separately.

Statistical analysis

A statistical analysis was conducted to determine the effects of *Cannabis* plant cultivation on cesium and strontium absorption. It should be considered that the ICP machine did not show any significant result based on the existence of cesium in the samples. Therefore, the two-sample test was used for strontium. The correlation coefficient test was used to examine the relationship between the concentration of strontium in the soil and the strontium uptake rate by the *Cannabis* plant.

Data were analyzed using the SPSS statistical package version 15.0 for Windows (SPSS Inc., Chicago, USA).The significance level was set at $P < 0.05$.

RESULTS

With regard to the cesium element, the ICP did not show any finding, that is, this element was not absorbed by *Cannabis*. The ICP measurements for strontium are presented in Table 2.

The control samples were given to the ICP, but the soil of these plants did not have the aforementioned elements, the negative answer was a confirmation by the machine.

As depicted in Figure 1, the roots of *Cannabis* have the maximum strontium absorption in comparison with the stems and leaves; and possibly the maximum absorption concentration by *Cannabis* is 60 ppm in the three main organs. If this strontium concentration increases, *Cannabis* strontium absorption will reduce, for example, *Cannabis* absorption in 60 ppm was 42, 35, and 23%, in the roots, stems, and leaves, respectively, but in 80 ppm it was 41, 39, and 20%.

Figure 2 shows that the *Cannabis* strontium uptake from the soil was not related to the soil concentration of the element.

Only two variables of strontium absorption and the main parts of the plant showed significant association. The percentage of strontium absorption was 45% in the roots, 40% in the stem, and the minimum absorption was in the leaves (15%).

The average of the two independent samples was defined [Table 3]. The first group consisted of 12 samples, containing

Table 1: Main characteristics of the most important radionuclides in the environment

Main occurrence	Isotope	Half-life (year)	Principal radiation
Natural and nuclear reactor	^{14}C	$5.7 * 10^3$	β-
Natural	^{40}K	$1.3 * 10^9$	β-
Natural reactor	^{90}Sr	28	β-
Natural reactor	^{134}Cs	2	β-, γ
Natural reactor	^{137}Cs	30	β-, γ
Natural reactor	^{239}Pu	$2.4 * 10^4$	α, χ-rays

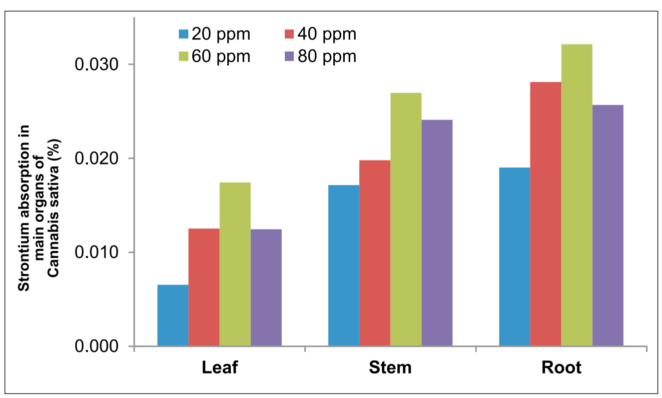

Figure 1: Strontium absorption in the main organs of *Cannabis Sativa* in different concentrations

Table 2: Inductively Coupled Plasma (ICP) measurements

Concentration (ppm)	Leaves (mg/l)	Stems (mg/l)	Roots (mg/l)	Leaves (mg)*	Stems (mg)	Roots (mg)
80	0.830	1.606	1.712	0. 1245	0. 2409	0. 2568
60	1.163	1.798	2.142	0. 17445	0. 2697	0. 3213
40	0.835	1.320	1.874	0. 12525	0. 198	0. 2811
20	0.436	1.144	1.268	0. 0654	0. 1716	0. 1902

Volume: 15 ml, *The amount of strontium (mg) in 1 g of dried samples

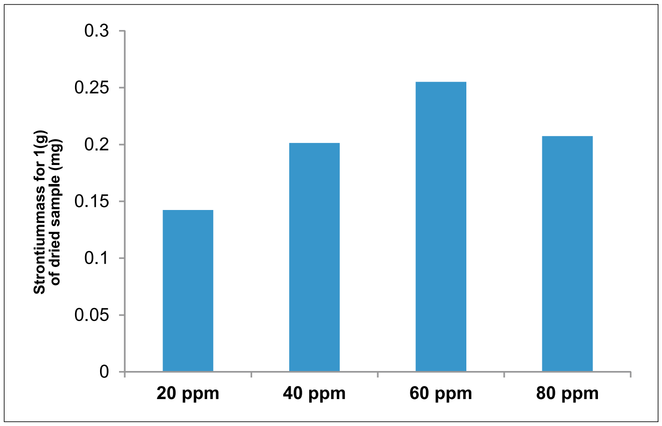

Figure 2: Amount of strontium uptake in *Cannabis* from different concentrations of strontium in the soil

Table 3: Evaluation of the relationship* between the strontium concentrations in soil and its absorption by *Cannabis*

	Concentration	Plant's organ	Strontium
Concentration			
r	1	0	0.387
P	_	1	0.214
n	12	12	12
Plant's organ			
r	0	1	0.795
P	1		0.002
n	12	12	12
Strontium			
r	0.387	0.795	1
P	0.214	0.002	_
n	12	12	12

*Pearson correlation; Correlation is significant at the 0.01 level (two-tailed)

different concentrations of strontium in the main organs of the *Cannabis* plant. The second consisted of three control samples (roots, stems, and leaves).

We did not find any relationship between the increasing of strontium concentration in the soil and increasing of the absorption amount by *Cannabis*.

DISCUSSION

Our findings suggest that strontium can be absorbed by *Cannabis sativa*, with most absorption done by roots, followed by the stems and leaves. However, cesium does not reach the plant because of its single capacity and inactive complex formation.[15] Considering that cesium has single capacity and strontium is a two-capacity element, in our study, *Cannabis* did not absorb single-capacity elements like cesium. This could be because cesium is the kind of element that produces inactive complexes, which become unavailable and non-absorbable by the plant. Moreover, we have documented that the plant root is the most active part of the *Cannabis* plant, and has the highest rate of strontium uptake.

Phytoremediation is still a new approach with a great potential for development. It needs widespread multi-dimensional knowledge of plant biology, agriculture, soil science, microbiology, and genetic engineering.

In general, the advantages and disadvantages of phytoremediation for each project should be assessed, to determine the suitability of this procedure. Combining the mentioned techniques provides a very high potential for effective phytoremediation in polluted sites. Moreover, it should be mentioned that after a constant period of phytoextraction the soil may lose its potential for later use. Therefore, soil nutrient management should be an essential part of phytoremediation, and this needs cautious consideration.

The limitations of phytoremediation are its long process and its restriction to low-depth sites. Moreover, the use of non-native and invasive plants may have an impact on the environment biodiversity.[16]

In this regard, bad weather is one of the most important concerns that should be considered, because it causes limited plant growth and reduces production of the biomass.

Thereafter, concentration of the soil contaminants should be considered, because in the presence of plenty of pollutants for toxic plants, successful phytoremediation would be reduced.[17]

General techniques for removing soil pollution are divided into two categories. The *ex situ* method needs removal of contaminated soils for treatment, in place or out of place, and returning of the treated soils to its main place. The *in situ* method does not need excavation for soil treatment.

Technologies of polluted soil treatment with this method are destroying or reforming pollutants, immovably, for reducing biological stress and separation of pollutants from the soil mass.[18]

Phytoremediation is inexpensive, stable, and suitable as an alternative to conventional methods reform, in developing countries is rapidly created. To achieve this goal, fast growing plants with high biomass and good quality are required.[18]

Phytoremediation experiments are performed more on a laboratory scale in hydroponic conditions and heavy metals have been added to them, however, the soil environment is quite different. In real soil, many metals are in dissolube form, their availability is low, and this is the greatest problem. Many plants that should be recognized are not known and efforts should be made to know more about their physiology.

CONCLUSION

Our findings suggest that *Cannabis sativa* can absorb strontium, with most absorption by the roots, followed by stems and leaves. However, cesium does not reach the plants because of its single capacity and inactive complex formation. These findings can be implemented in environmental protection issues.

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