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REGULAR ARTICLE

CAPABILITY OF LEMNA GIBBA TO BIOSORB CESIUM-137 AND COBALT-60 FROM SIMULATED HAZARDOUS RADIOACTIVE WASTE SOLUTIONS

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ABSTRACT

Low and intermediate level radioactive wastes are generated from daily applications of radioisotopes in various medical, industrial, agricultural and research fields. The release of these wastes to the surrounding environment represents a major complicated ecological crisis. Treatment of that radwastes is the most essential process in the radioactive waste management scheme. For simplicity and low running costs, phytoremediation technique has been candidate for processing some selected hazardous liquid waste streams. The submitted work aims at evaluating the capability of one of the aquatic plant, namely *Lemna gibba*, to biosorb Cs-137 and Co-60 from aqueous radioactive waste simulate. The study discusses, in batchwise laboratory scale experiments, the parameters that may affect the efficiency of *Lemna gibba* to bioremove and bioaccumulate the two radionuclides (e.g: contact time, pH value and the initial activity content of the waste simulate, light effect, biomass used, . . .). The uptake values, biosorption efficiency percentages, rate constant and isotherm factors were evaluated for the process. The uptakes values for Co-60 and Cs-137 respectively recorded 1213 Bq/gm and 872 Bq/gm from the waste simulate solution containing 6100 Bq and at pH=6.9 after 24 hours contact time. The results obtained exhibit the potential of the wide aquatic plant Lemna gibba to be used as a biological sorber for cesium-137 and cobalt-60 from their low and intermediate level aqueous radioactive waste stream successfully and efficiently.

Keywords: Radioactive waste solution, Cs-137, Co-60, *Lemna gibba*, Phytoremediation

INTRODUCTION

Protection of the environment from the hazardous radioactive wastes pollution is an era of study which still commands considerable attention to optimize the vast usage of nuclear technology for the everyday needs. Radioactive wastes can be classified based on their physical state to gaseous, liquid and solid wastes.

The primary sources of aqueous radioactive wastes in a country without nuclear fuel cycle activity, like Egypt, are nuclear research, production and application of radioisotopes, decontamination and decommission of nuclear installation. In general as a result of these activities more than 75% of aqueous low and intermediate level radioactive wastes are generated **(IAEA, 1984)**.

Treatment of radioactive wastes includes operations intended to benefit safety and economy by changing the waste characteristics. The three main basic treatment process objectives are: waste volume reduction, removal of dangerous radionuclides and change in physical state and/or chemical composition of the radwaste.

Bioremediation is a natural process and is, therefore, perceived by public as an acceptable radioactive waste treatment method to replace the conventional techniques based. Biosorption is considered to be a fast physical and/or chemical processes and assumed route for plant to clean the waste streams from their hazard radiocontaminants. According to literature biosorption can be divided into two main processes: adsorption of the ionic species on the cell surface and bioaccumulation within the cell **(Mata et al., 2009)**.

The ability of aquatic plants to sorb and accumulate metals from their aquatic environment has been demonstrated by a number of researchers **(Zurayk et al., 2002; Keskinkan et al., 2004; Mkandawire and dudel, 2007; Kamal et al., 2007)**. On the other hand, rare works exist in the literature on the phytoremediation capacity of plant for the radioactive contaminants.

The objective of this study is to evaluate the potential of the nominated wide *Lemna gibba* aquatic plant as a phytoremediation agent in the concept of aqueous radioactive waste treatment. The parameters that measure the extant of radiocontaminants (e.g: Cs-137 and Co-60) biosorption were comparatively assessed (e.g: biomass content, radioactivity concentration, pH of waste streams, …) and the results reached were reported precisely.

MATERIAL AND METHODS

Materials

Simulated radioactive waste stream

Carrier free solutions of Cs-137 and Co-60 were used to spiked definite volumes of tap water to have gradient increases in activity contents streams simulating the radioactive waste.

Radioisotopes

Carrier free solutions of Co-60 ($CoCl_2$ in 0.1M HCl) and Cs-137 (CsCl in 0.1M HCl) with original specific activity 115 M Bq/ml and 127 M Bq/ml, respectively were purchased by Radioisotope Center Polatom, Institute of Atomic Energy, Poland.

Plant

Lemna gibba (duck weed) is a free floating aquatic plant from family *Lemnaceae*. They commonly grow in stagnant or slow flowing water throughout tropical and temperate zones. They were collected from El-Mareotea Canal in Giza district near Cairo and identified at the herbarium of the National Research Center. For the current work, the wild *Lemna gibba* was used for treatment of the prepared simulated radioactive waste solutions.

Methods

Batch studies were carried out, for a predeterminate period of time in laboratory environment using glass containers each having 50ml aqueous radioactive waste simulate. Every container was charged by predetermined fresh weight of *Lemna gibba*. Crab samples of the waste simulate before starting the phytoremediation process were withdrawn and analyzed radiometrically using "3X3" well type NaI crystal (efficiency 85%) based on Multichannel Analyzer PCA-P-USA. After the addition of the *Lemna gibba* to the container, samples from the treated media were collected at definite time intervals and their radioactivity contents were also counted.

Some parameters that supposed to affect the biosorption efficiency of *Lemna gibba* for the both radionuclides Cs-137 and Co-60 (e.g: contact time, radioactivity content, pH of the simulated waste solution, the biomass of the plant added,…) were assessed systematically.

Data evaluation

To evaluate the phytoremediation process the amount of the radioactivity uptake within the biomass of *Lemna gibba* was calculated as follows:

$$q = (a_o - a_t) \times (V/M).$$

Where:

q: activity uptake (Bq/gm).

a_o: the initial activity (Bq/ml).

a_t: the final activity content after t period (Bq/ml).

V: volume of waste solution (ml).

M: mass of the plant added (gm).

Also to rank the capability of the plant to bioaccumulate both Cs-137 and/or Co-60 from simulate radioactive waste stream, biosorption efficiency percent was calculated according to the coming relation:

$$R = ((a_o - a_t)/a_o) \times 100.$$

Where:

R: the biosorption efficiency percent.

Biosorption models

Isotherm studies

Langmuir isotherm is applied to identify the interaction of the radionuclide species and the sorbent biomass. The following represents the linearized Langmuir isotherm equation:

$$q_t/q_{max} = ba_t/(1+ba_t)$$

Where:

q: experimental uptake at time t (Bq/gm).

q_{max}: maximum radioelement uptake (Bq/gm).

b: a constant related to the affinity of binding site (ml/Bq).

To fit Langmuir equation, the relation **a_t/q** versus **a_t** was plotted for both Cs-137 and Co-60 biosorption **(Melcakova and Ruzovic, 2010)**.

Separation factor

The essential characteristics of the Langmuir isotherm can be expressed in terms of separation factor R_L which is defined as:

$$R_L = 1/(1+ba_o)$$

It is known that R_L values between 0 and 1 indicate favorable adsorption **(Mekay et al., 1982)**.

Kinetic studies

Sorption data for Cs-137 and Co-60 by *Lemna gibba* were described based on the intraparticle diffusion model that can be expressed as:

$$q_t = k_p (t)^{1/2} + c$$

Where:

k_p: the intraparticle diffusion rate.

t: the time interval (h).

c: constant.

This model applies when a linear representation of q_t various $(t)^{1/2}$ is obtained, and k_p can be calculated from the slope of the curve **(Weber and Morris, 1962)**.

RESULTS AND DISCUSSION

Lemna species have many unique properties ideal for remediation process among them, they are fast grown and primary production, have high bioaccumulation capacity and a good phytoremediation agent **(Körner and Vermaat, 1998)**.

Effect of contact time on uptake and biosorption efficiency

The behavior of *Lemna gibba* as bioaccumulator performer for radiocesium and/or radiocobalt as a function of contact time up to 3 days was study by growing a definite biomass of the plant in certain volume of simulated radioactive waste stream spiked with the both radionuclides. The data obtained are represented in fig (1) for the uptake (a) and the biosorption efficiency of the plant in percent (b), respectively.

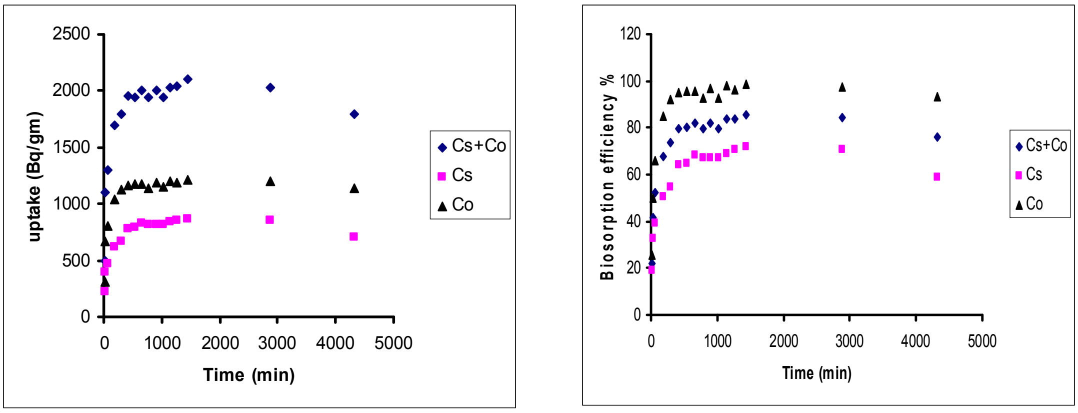
It is clear from the data obtained that the biouptake of the radioactivity increased by increasing the contact time under the lab environment temperature (25±5Ċ). The biouptake reached maximum after 24 hours. By expanding the contact time a decrease in uptake was observed.

The behavior of the *Lemna gibba* during phytoremediation process can be shown in fig (1-b). It could be distinguished that the capability of plant to biosorb Cs-137and/or Co-60 increases till 24 hours. After this time the radiocontaminants were back released to the solution medium. This may be attributed to exposure of plant to gamma irradiations from

^{137}Cs (E=0.662 Mev) and ^{60}Co (E=1.17 Mev and E=1.33 Mev). These high radiation energy affects negatively the habits plant living cell and leads to the absolve of the radioactivity to the solution again.

The main radiotoxicty symptoms such that stunted growth, chlorosis of leaves with reddish brown spots that can be followed by necrosis of the plant tissue were observed following the exposure. Therefore, simultaneously substantial active effluxing possibly through the plant tissue and by time leaching of radioactivity from the necrosed tissue can be the key mechanism as the activity content in the simulated aqueous waste drastically increased after the first 24 hours. Similar trend was described by **JayaWeera et al. (Jayaweera et al., 2008)**.

Based on the results reached it could be stated that, the optimum time for the treatment process is near 24 hours and after that the plant should be removed from the media and treated as solid waste and fresh batch of plant can be added to continue the phytoremediation function.

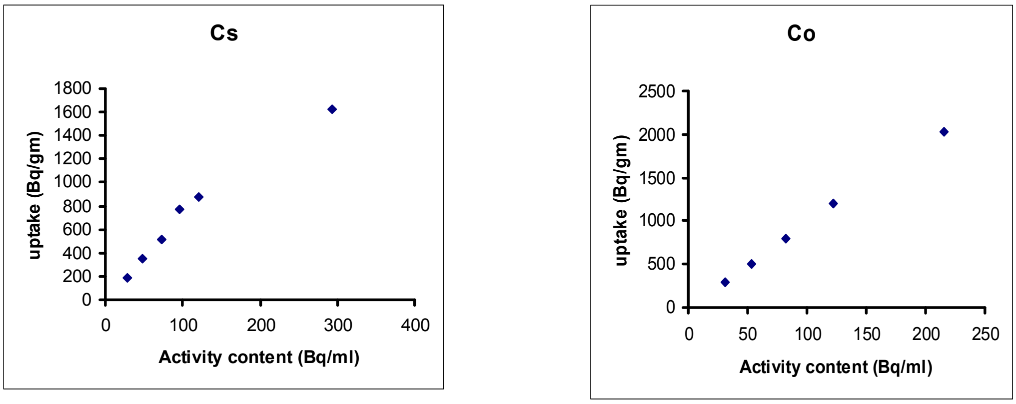


(a) **(b)**

Fig 1 the biouptake value (a) and the biosorption efficiency (b) of *Lemna gibba* for Cs-137 and Co-60 mixture as a function of contact time

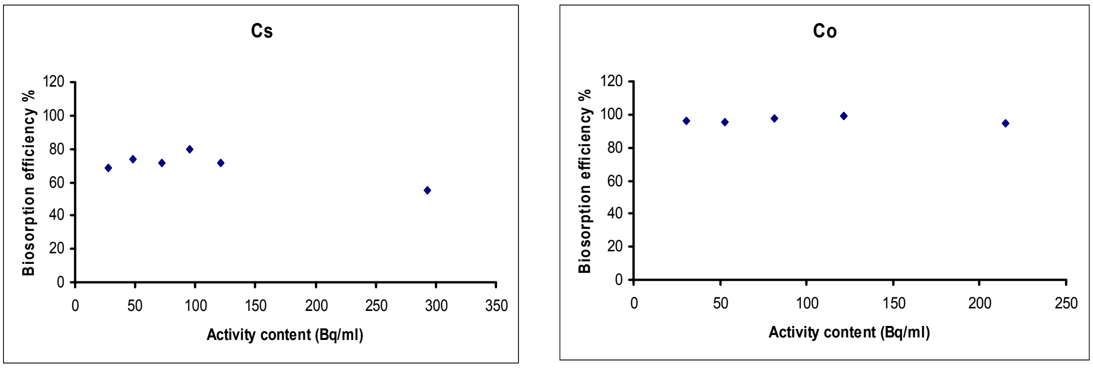
Biosorption efficiency and uptake as a function of initial radioactivity contents

The effect of the initial activity content of both Cs-137 and Co-60, found in mixture, on their uptake values and plant biosorption efficiency percentages was studied in the range of (27.8 Bq/ml) to (293.4Bq/ml) for radiocesium and of (30.6Bq/ml) to (215Bq/ml) for radiocobalt, respectively. Laboratory batch experiment was carried out at pH value=6.9 and at contact time 24 hours using 5gm fresh *Lemna gibba* plant, and the data obtained were shown in fig (3) and (4). It is clear that, the increase in uptake values, for the two radioisotopes, are directly proportional to the initial radioactivity content added. This relationship is probably attributed to the higher interactions between the radioelement (i.e: Cs^{+} and Co^{2+}) and the biosorbent sequestering sites, fig (3). Consequently the increase in the radioactivity contents would compete for the available binding sites in the biosorbent surface that ready for accumulating both Cs^{+} and/or Co^{2+} at the higher radioactivity levels. On the other hand, increasing the initial activity contents of Co-60 added to the constant weight of *Lemna gibba* shows undetectable effect on the biosorption efficiency of the plant for bioaccumulating the radionuclide, Fig (4). While increasing the initial content of Cs-137 accompanied by gradual increasing in the biosorption efficiency percentage and continuing increase in the initial radiocesium contents added results in drop of biosorption efficiency. The obtained data could be explained on the basis that: the biosorption efficiency of *Lemna gibba* for ^{60}Co is independent on the initial activity content because cobalt is one of the essential elements needed for the plant **(Kamel et al., 2007)**. For Cs-137 it should be notified that, 100Bq/ml is considered threshold value and beyond this figure the biosorption efficiency of the plant decreased. This may be attributed to the high radiotoxicty of Cs-137 to the plant **(White and Broadley, 2000)**.



(a) (b)

Fig 2 The effect of initial radioactivity contents on the biouptake of ^{137}Cs (a) and ^{60}Co (b) from aqueous radioactive waste simulate



(a) (b)

Fig 3 The effect of initial radioactivity contents on the biosorption efficiency of *Lemna gibba* for Cs-137 (a) and Co-60 (b)

Effect of pH value of the waste solution simulate on biosorption efficiency

The dependence of *Lemna gibba* biosorption efficiency on the pH values of the simulated radioactive waste solution under treatment was investigated in a wide range starting from the acidic (pH=2.9) to the alkaline (pH=10.9) based on the removal percentages of Cs-137 or Co-60 from their waste streams. The data in fig (4) indicated that different behavior of the aquatic plant toward radiocobalt and radiocesium removal. As seen clearly that the biosorption efficiency of *Lemna gibba* to ^{60}Co not affected by the change in pH values of the medium and nearly the plant can bioaccumulate the radiocobalt at the wide range of pH values tested. That may referred to that cobalt is one of the essential metals required by the plant for the growth and development as previously stated and, hence, they have the ability to accumulate it at wide range of pH values **(Jadia and Fuelkar, 2009)**. In addition most plants and microorganism use cobalt as a primary source for vitamin B12 production **(Moore, 1991)**. On the other hand the biosorption efficiency for ^{137}Cs clearly affected by the pH values of the treated waste stream. It is clear that the ability of the aquatic plant increased gradually by raising the pH values of the growing media to record the optimum biosorption efficiency percent at pH near 7. Slight lower affinity to radiocesium at the acidic medium can be referred to the competition between the monovalent (Cs^+ cation) and the protons (H^+) for sorption binding sites on the cell wall of the plant and consequently the concomitant decrease in negative charge in the cell surface. Similar trend was described in pervious published works. As the pH values increased the surface charge on the cells become negative and the biouptake of radiocesium increased and this suggested that the positively charge Cs^+ can be binded through electrostatic attraction to the negatively functional groups on the surface of biosorbent. It has been assumed that the reduction in bioaccumulation of Cs-137 as the pH increased beyond its optimum values may be referred to the high solubility of cesium at the alkaline end.

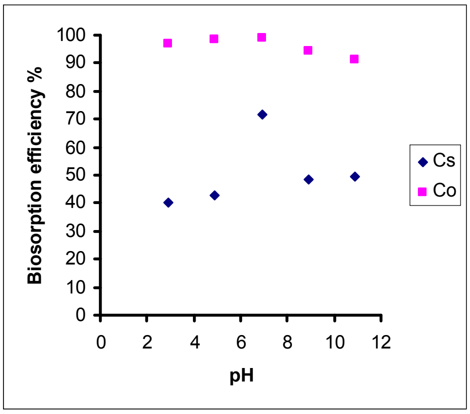
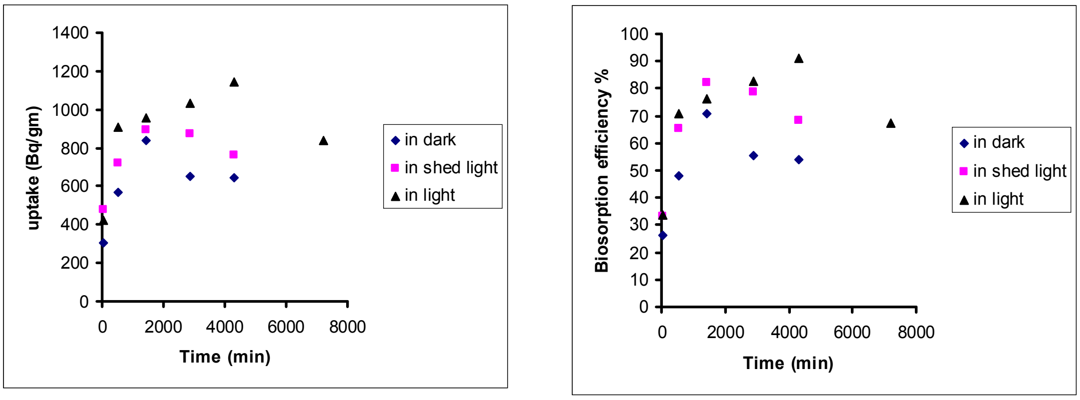


Fig 4 The effect of pH value of waste solution simulate on the biosorption efficiency of *Lemna gibba* for ^{137}Cs and ^{60}Co

Effect of light on biosorption efficiency and uptake

Most *Lemna* species are relatively insensitive to environmental change e.g: pH, temperature and availability of resources including photosynthetic radiation **(Mkandawire and Dudel, 2007)**. Three batch culture modes were prepared by adding 5gm of *Lemna gibba* to each glass container containing simulated radioactive waste solution with initial activity content (~250Bq/ml). The first batch was grown under direct sun light, the second was left at the shed light of the laboratory while the last one was put in dark place. The remain total radioactivity in the solution at different time intervals for the three batches were followed radiometrically and the data obtained were represented in fig (5). It is clear that, for the plant grown at direct sun light, the maximum biosorption efficiency percent for the removal of radioactive contaminants reaches later (t_{max}=72hs) compared to that grown at the shed light or under darkness which recorded nearly the similar t_{max} (~24hs). Same trends were observed for the uptake values for Cs-137 and Co-60, where in direct sun light, they recorded slightly higher values after 72 hours compared to experiments performed under shed light and dark conditions. It is worth mention that in the direct sun light, the plant shows detectable resistance to the backrelease of the two radionuclides and postpones it to the 5th day compared to the 3rd day in the experiments carried out at shed light and darkness, Fig (5). This data could be explained on the basis that: there is an intimate relation exists between the photosynthesis and the mechanism of the radioactive bioaccumulation. Where the plant in sun light is having a rapid growth and vegetative reproduction rates due to the ultraviolet radiation resulted in fast profilteration which effectively increased the bioaccumulation of the radiocontaminants **(Mkandawire and Dudel, 2007)**. On the other hand, at darkness, suppression at the rate of the plant bioactivities may persist.

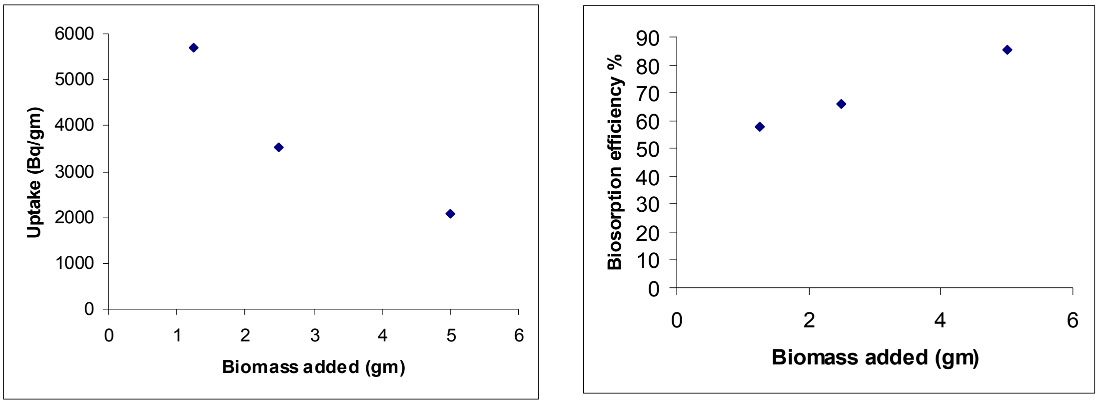


(a) **(b)**

Fig 5 The effect of light on the biouptake (a) and on the biosorption efficiency (b) of *Lemna gibba* for Cs-137 and Co-60

Uptake of radiocontaminants and biosorption efficiency as a function of wild *Lemna gibba* biomass added

Physicochemical mechanism for the removal of radiocontaminants from their waste streams which may be compared by the general term biosorption includes adsorption, ion-exchange and entrapment which are features of living biomass **(White et al., 1995)**. The effect of the amount of the fresh biosorbent that added to constant volumes of the simulated radioactive solutions containing the same initial total activity content at pH=6.9 on the biouptake values and the biosorption efficiency percentages was study at batchwise experiments and data reached were represented in fig (6).



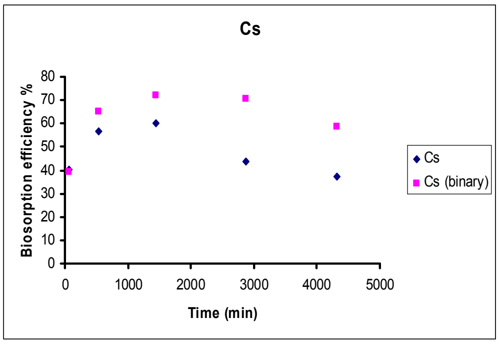
(a) **(b)**

Fig 6 The effect of biomass added on the biouptake (a) and on the biosorption efficiency of *Lemna gibba* for ^{137}Cs and ^{60}Co removal

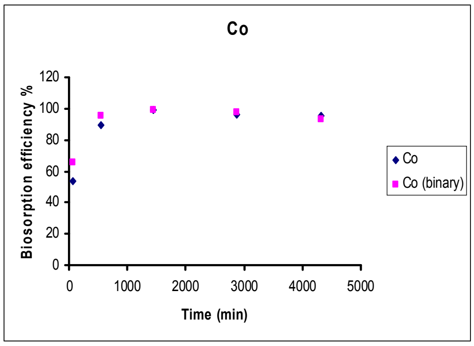
It is clear that increasing the added biomass enhances positively the efficiency of the *Lemna gibba* to bioaccumulate radiocontaminants from simulated waste streams. As it is a floating plant, it forms a dense mat cover the solution surface implies its ability to survey up a wide area for quick removal of the radionuclides. On contrary, the increase in the biomass dosage to the constant radioactive content accompanied with a downturn in the uptake of radionuclides. This decline trend may be attributed to the unsaturation in the active sites due to their overlapping as a result of increasing the amount of biomass used.

Effect of the type of radionuclides on the biosorption efficiency of *Lemna gibba*

Radiowaste solution streams contaminated with radionuclides particularly ^{137}Cs and ^{60}Co pose a long term radiation hazard to human health through expose via the food chain and other pathways under uncontrolled releases of these wastes to the environment. The effect of the type of radionuclide on biosorption process from waste streams spiked with the both radioisotopes in single or in binary composition was study. Laboratory batch phytoremediation experiments were carried out for three aqueous radioactive waste simulated solution spiked with Cs-137, Co-60 or the both radionuclides at pH=6.9 for time intervals up to 72 hours. The biosorption efficiency percentage for each batch experiment was calculated and the results reached were represented in fig (7). The biosorption efficiency of *Lemna gibba* for cobalt-60 is not affected by the presence of cesium-137 in the same solution, while the availability of Cs-137 to the plant is affected by the existence of Co-60. This corroborated that many of the functional groups present on the cell wall and membrane are nonspecific and different cations compete for the binding sites and plant specific factors are the main sources for biosorption of radiocobalt compare to radiocesium. Similar trend was reported by **Gerzabek et al. (Gerzabek et al., 1998)**.



(a)



(b)

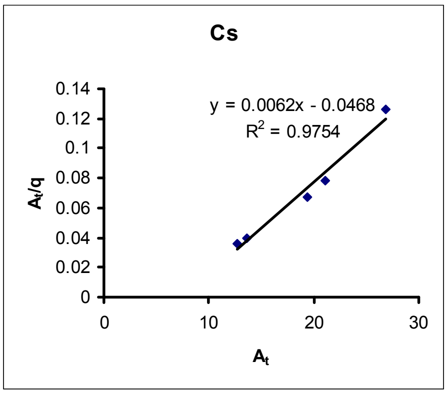
Fig 7 The effect of type of nuclides on the biosorption efficiency of *Lemna gibba* for Cs-137 (a) and Co-60 (b) from waste solution simulates

Biosorption models

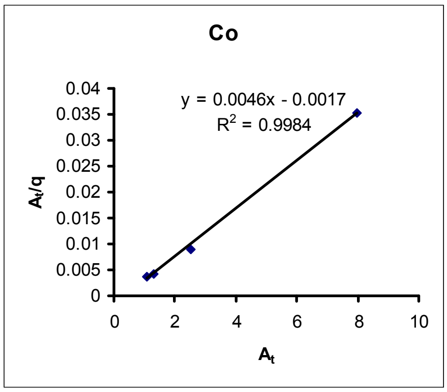
The mechanisms of biosorption of radionuclides are understood only to a limit extent and the uptake of them by *Lemna gibba* in batchwise systems seems to occur in two stages: an initial rapid one, marked by passive uptake, followed by much slower energy-dependent active process. The first stage is mainly physicochemical process takes place at the surface of the biosorbent cells. The sorption process includes the transport of the radiocontaminantes from the bulk solution to the exterior surface of the plant where the active sites are found **(Salehi et al., 2008)**.

Isotherm study

In this study, the Langmuir isotherm model was applied to identify the interaction of the radionuclide species and the sorbent biomass. The data is represented in fig (8). Langmuir isotherm assumes that sorption occurs uniformly on the active sites of the sorption, and once sorbate occupies a site, no more sorption can take place at this site **(Ho et al., 2002)**. The best fit of equilibrium data for both radiocesium and radiocobalt for Langmuir expressing confirms the monolayer coverage of each radioelement onto *Lemna gibba* biomass. The R^2 values for both radionuclides were very close to 1, which revealed the extremely good applicability of the Langmuir model to this adsorption process.



(a)

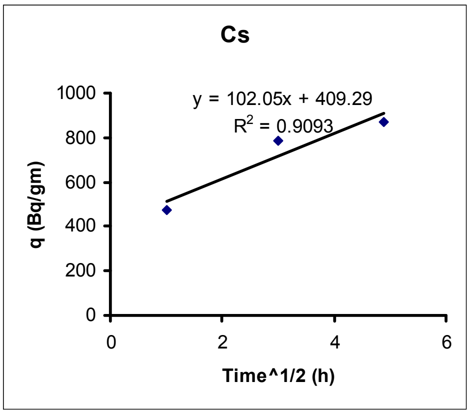


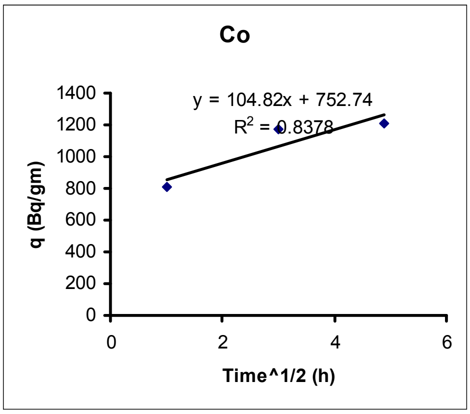
(b)

Fig 8 The linearized Langmuir adsorption isotherm for ^{137}Cs (a) and ^{60}Co (b) from aqueous waste solution simulate based on *Lemna gibba*

Kinetic study

Biosorption kinetics can be fit to the intraparticle diffusion model which proposed by Weber-Morris. This model assumed a two-step biosorption process-metal binding to the biomass surface, followed by metal diffusion through its pores **(Gerzabek et al., 1998)**. It's clear from fig (9) that diffusion was involved during biosorption, but was not only the rate controlling step since the linear plots did not intercept the origin. This also indicates that, other processes including ion-exchange or entrapping, as previously state, are sharing **(Weber and Morris, 1962; Demrbas, 2008)**. However to understand how the two radionuclides are sorbed by *Lemna gibba*, it is essential to identify the functional groups on the cell wall responsible for that binding. Charged and polar functional groups on the protein surface and phenolic compounds may be primarily involved in the radiocontaminants removal. These groups have the ability to bind the radionuclides by replacement of hydrogen ions for metal cations (i.e. Co^{2+} or Cs^{+}) or by donation of an electron pair to form metal complex **(Lavecchia, 2010)**.





(a) **(b)**

Fig 9 The intraparticle diffusion model for ^{137}Cs (a) and ^{60}Co (b) sorption from aqueous waste solution simulate

Table 1 Kinetic and Langmuir parameters for both Cs-137 and Co-60 based on biosorption by *Lemna ginbba*

Parameters	**Cs-137**	**Co-60**
Initial activities (Bq/ml)	121.3	122.7
Biomass added (gm)	5	5
pH	6.9	6.9
k_p	102.05	104.82
R^2	0.9093	0.8378
b	0.13	2.71
R_L	0.06	0.003

The Kinetic and Langmuir data represented in table (1) confirmed the high affinity of *Lemna gibba* to bioaccummulate both radiocesium and radiocobalt from their waste solution streams.

CONCLUSION

Lemna gibba represents a promising biosorbent that candidate to bioseparate and bioaccumulate two risky gamma emitters, namely, cesium-137 and cobalt-60 from low and intermediate radioactive waste solutions generated from peaceful applications of nuclear technology in the daily life. The proposed batchwise phytoremediation process reached maximum within approximately 24 hours at pH=6.9 and under laboratory conditions (e.g: room temperature, shed light and atmospheric pressure). The uptake values of the two radionuclides from their waste streams by *Lemna gibba*, under the state conditions, were 1213 Bq/gm and 872 Bq/gm for Co-60 and Cs-137 respectively. The process is environmentally non-destructive and cost-effective. Therefore, the destruction or harvesting of *Lemna gibba* biomass in surface water should be avoided where it can be applied as phytoremediation agent for various radioactive contaminants as well as other hazardous materials. Besides the low running cost advantage, the process is very simple and, therefore, can be smoothly applied at semifield and field levels.

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