Chemosphere 215 (2019) 15–24

Contents lists available at ScienceDirect

Chemosphere

journal homepage: www.elsevier.com/locate/chemosphere

# Mechanisms of strontium's adsorption by *Saccharomyces cerevisiae*: Contribution of surface and intracellular uptakes

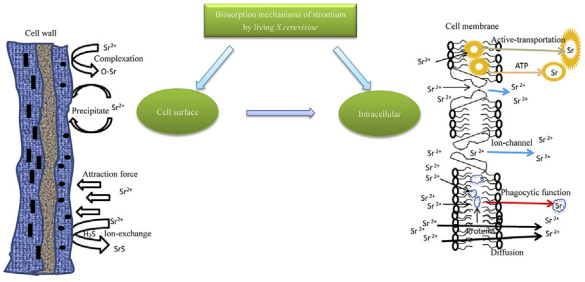
Liang Qiu, Jundong Feng*, Yaodong Dai, Shuquan Chang

*Department of Materials Science and Technology, Nanjing University of Aeronautics & Astronautics, Nanjing 210016, PR China*

## HIGHLIGHTS

- Yeast cells bind strontium from solution via surface and intracellular processes.
- Functional groups participated in the process are explored by chemical blockage.
- Ion-exchange and complexation are the main mechanisms in surface sorption.
- Metabolically-dependent proteins may play an important role in bioaccumulation.

## GRAPHICAL ABSTRACT




## ARTICLE INFO

*Article history:*
Received 9 May 2018
Received in revised form
17 September 2018
Accepted 28 September 2018
Available online 1 October 2018

Handling Editor: Martine Leermakers

*Keywords:*
$Sr^{2+}$ Bisorption
Plasmolysis treatment
Desorption
Chemical blockage
Protein analysis

## ABSTRACT

The objective of this work was to explore the mechanisms participating in strontium sorption by living *Saccharomyces cerevisiae* (*S. cerevisiae*). The location of strontium adsorbed by *S. cerevisiae* was studied by our plasmolysis treatment. The contribution of physical and chemical mechanisms was determined quantitatively by desorption and blockage of functional groups. Moreover, our results indicated that bioaccumulation also played a major role in biosorption by living cells. Thus, supplementary methods including 2-DE (two-dimensional electrophoresis) and Matrix-Assisted Laser Desorption/Ionization Tandem Time of Flight Mass Spectrometry (MALDI-TOF-TOF) were employed to analyze the different proteins.

The subsequent desorption % of $Sr^{2+}$ by Distilled Water (DW), $NH_4NO_3$ and EDTA-$Na_2$ from $Sr^{2+}$ loaded sorbents indicated a minor role for physical adsorption, while ion exchange and complexation were responsible for approximately 20% and 40%. Specific blockage of functional groups revealed that carboxyl and amine groups played an important role in $Sr^{2+}$ binding to the living *S. cerevisiae*. From our MALDI-TOF-TOF results, we concluded that 38 proteins showed up-regulated expression profiles and 11 proteins showed down-regulated after biosorption. Moreover, proteins belong to: phagocytic function (Act1p); ion channel (S-adenosylmethionine synthase); glycolysis (Tubulin) may directly involve in strontium bioaccumulation.

In conclusion, the present work indicates that the strontium sorption mechanism by living *S. cerevisiae* is complicated including ion-exchange along with complexation as the main mechanism, whereas the other mechanisms such as physical adsorption play a minor contribution. Metabolically-dependent proteins may play an important role in bioaccumulation.



* Corresponding author.
*E-mail address:* jundongfeng@nuaa.edu.cn (J. Feng).

https://doi.org/10.1016/j.chemosphere.2018.09.168
0045-6535/

## 1. Introduction

Nowadays, there has been a growing concern on the aquatic environment contamination because of the large amounts of radioactive waste generated by human activities (such as storntium-90). Biosorption method, as a new emerging method for radionuclides disposal, has shown the advantages of high sorption capacity, low production costs and less secondary sludge (Hans-Cur, 1995; Saeid et al., 2013). Moreover, in our previous studies, we found that the advantage of biosorption by living *S. cerevisiae* were more obvious because its rapid proliferation, high tolerance to waste levels, and excellent uptake ability of heavy metals and strontium (Qiu et al., 2017). However, the mechanisms whereby the living microorganisms that could cause the radionuclides removal from water are poorly understood.

Two basic mechanisms by which living microorganisms can remove radionuclides are (1) adsorption onto cell surface; (2) intracellular uptake and accumulation of radionuclides. In performed mechanisms studies, many researchers only focused on adsorption onto cell surface. The classification of the surface mechanisms includes: physical adsorption, ion exchange, complexation. Physical adsorption is the results of precipitation and van der Waals force. Such attraction force leads to adsorption of the nuclide on the surface of the cell. For ion exchange, it is defined as equal replacement of light metal ions in solid phase by metal ions in solution (Petrović et al., 2017). Complexation can occur directly by interaction of nuclide with anionic cell wall functional groups or owing to the phosphate moiety (Prakash et al., 2013).

For intracellular uptake and bioaccumulation (living), few researchers pay attention it since this process is more complex than adsorption and requires metabolic activity of cells (Shukla et al., 2017). It can be classified into: Biotransformation (Hullebusch et al., 2005); Transport mechanisms (Gadd, 1996) and Biosolubilisation (Gadd et al., 2002). Biotransformation is a chemical process. Microorganism which is actively growing possess the ability of biotransformation via redox reactions. This reaction can lead to formation of oxides, coprecipitates, ionic, organic or inorganic complexes of radionuclides (Francis and Dodge, 2009). The reduction of $Np^{5+}$ by *Desulfovibrio desulfuricans* has been reported by Icopini (Icopini et al., 2007). Transport mechanisms include: ATP-binding cassette transporters; metal channel and efflux transporters. It is attributed directly to the presence of the respective proteins transporter or the nuclide on permeability of the cell membrane (Jabbar and Wallner, 2015). Biosolubilisation is catalyzed by a variety of microorganisms in natural and engineered environments. It is a result of autotrophic metabolism during which, radionuclides are leached out from their solid matrices (Gadd et al., 2002). This part is affected by the oxidation state of the nuclide, pH, microbial activity (Kaksonen et al., 2017). Large-scale biosolubilisation has been mainly used for $Cu^{2+}$ (StefaNescu et al., 2010), $Co^{2+}$, $Ni^{2+}$, $Zn^{2+}$ (Yin et al., 2009), $Fe^{3+}$ (Constantinescu et al., 2013) through oxidative bioleaching, whereas it is yet to be implemented at industrial scale.

As we mentioned above, due to the sophisticated biosorption process, it is difficult to understand the biosorption processes using a singular mechanisms. Therefore, to further study the biosorption mechanism, it is necessary to adopt a comprehensive study including the contribution of surface and intracellular along with its reaction concurrently. Such studies enables us to facilitate to the development of potentially applicable biosorbents for wastewater treatment.

The aim of this article is (1) to identify quantitatively the intracellular and cell surface proportions of strontium ions by plasmolysis treatment. (2) To investigate the contribution of the involved mechanisms by desorption method: using deionized water (DW), $NH_4NO_3$ and EDTA-$Na_2$. (3) To assess the complexation mechanism by chemical pretreatment (blockage) of the functional groups. (4) For the first time using MALDI-TOF-MS to explore the different proteins between no-sorption $Sr^{2+}$ and sorption $Sr^{2+}$.

## 2. Material and methods

### 2.1. Microorganism and materials

*S. cerevisiae* (CICC 30225) cells were obtained from the China Center of Industrial Culture Collection (CICC). In our previous studies (Qiu et al., 2017), we had used cyclic irradiation method to culturing an irradiated S. *cerevisiae*: Y-7. In this paper, we will use Y-7 as living biosorbents and do our further researches. All chemicals were of analytical grade unless otherwise stated.

### 2.2. Biosorption experiments

The sorption process was carried out in batch mode, at constant temperature of 30 °C and shaking rate of 150 rpm. The samples of 10 g $L^{-1}$ sorbents were added into 10 mL of $Sr(NO_3)_2$ ($Sr^{2+}$ is used as a surrogate for $^{90}Sr$) solution with an initial concentration of 20, 50, 100, 200, 400 mg $L^{-1}$ at pH 7 in a 50 mL Erlenmeyer flask. 0.1 M HCl and 0.1 M NaOH solutions were used for pH adjustments. Based on our previous work (Qiu et al., 2017), $Sr^{2+}$ sorption reached a maximum state until 30 h for all cases and control experiments without biosorbents were also performed. After maximum attainment, the solution was separated from the sorbents by centrifugation (4000 rpm, 10min, 4 °C) and 5 mL of the supernatant were analyzed by Atomic Absorption Spectroscopy (AAS, Persee TA5-990, China) for the residual metal ions in the solution. The details of AAS are summarized as follows: Hollow cathode lamp: Strontium, Sensitive line: 460.7 nm; Operating current: 3.0 mA; Slit: 0.4 nm; Acetylene flow: 2000 mL $min^{-1}$; Burner high: 6.0 mm; Energy: 100%. The lamp were be preheated for 0.5 h before testing, the characteristic wavelength were corrected to 460.7 nm and the concentration of strontium was calculated (AA win program 3.0) by the absorbance of supernatant. Each supernatant repeated in triplicate. The verification regulation for AAS were according to JJG 674-2009 (S.1) and the quality controls were conducted by our program. (upper and low limit; error correction; Sensitivity correction; etc).

The amount of metal ion sorbed in equilibrium was expressed as adsorption capacity $q_e$ (mg $g^{-1}$) and calculated according to the following equation:

$$q_e = \frac{(C_o - C_e)V}{m} \quad (1)$$

Where $C_o$ and $C_e$ (mg $L^{-1}$) are the initial and equilibrium metal ion concentrations, respectively; V (L) is the solution volume and m (g) is the weight of wet sorbent. All experiments were conducted in duplicate.

### 2.3. Plasmolysis treatment

The thickness of *S. cerevisiae* wall is 0.1–0.3 μm, and it is mainly consisted by D-dextran, D-mannan and a small amount of proteins, fats. Based on previous study (Mclellan et al., 2010), in this paper we used physical mechanics method to break the cell wall.

After sorption experiment conducted as described above, the biomass was centrifuged and recovered. 0.5 g prepared strains from each strontium metal concentration were suspended in 1 g glass beads and 0.5 mL of concentrated hydrochloric acid. The reaction mixture was shaken on a High-speed thermo shaker (2000 rpm) for