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Bioremediation of Cr(VI) Polluted Wastewaters by Sorption on Heat Inactivated *Saccharomyces cerevisiae* Biomass

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ABSTRACT: The potential of heat inactivated *Saccharomyces cerevisiae* in the bioremoval and reduction of Cr (VI) ions from wastewaters was evaluated in terms of metal uptake in time and at equilibrium, and biosorption efficiency, by varying pH, biosorbent doses, contact time and temperature, in batch mode. During the sorption process, the heat inactivated biomass of the yeast *Saccharomyces cerevisiae* is capable of reducing Cr(VI) to Cr(III). Different kinetic models based on adsorption and reduction are used to represent the kinetic data of Cr(VI) bioremoval by *S. cerevisiae,* in explaining the biosorption mechanism of heavy metals and potential rate-controlling steps, in the perspective of full-scale process design. The results indicated some potential differences in the Cr(VI) removal mechanism at different experimental conditions. FTIR and SEM analysis were performed as well as to elucidate the mechanism of metal bioremoval by *S. cerevisiae.* FTIR spectra indicate that heavy metal bioremoval process doesn't imply in this case the formation of stable covalent bonds, but it is predominantly based on chemical interactions, ion-exchange type. The SEM micrographs of Cr-loaded yeast, indicates that the surface morphology doesn't change much after chromium ions were uptaken. This leads to the conclusion that Cr(VI) reduction occurs at the interface of the adsorbent.

Key words: Biosorption, Cr(VI) reduction, Yeast biomass, Kinetics, Sorption mechanism

INTRODUCTION

The increase in environmental contamination is a major consequence of industrial development, with a particular emphasis on pollution with heavy metals. Unlike many organic pollutants, which eventually degrade to carbon dioxide and water, most metals are not destroyed, but they are accumulating in the environment, especially in lake, estuarine, or marine sediments at an accelerated pace (Sarkar, 2002; Cvijovic *et al.*, 2011; Cozma *et al.,* 2010). Metals can also be transported from one environmental compartment to another, which complicates the containment and treatment problems (Jackson *et al.,* 2010; Pavel *et al.*, 2010; Wuana and Okieimen, 2011). Consequently, heavy metals are closely connected with environmental deterioration and the quality of human life, and thus have aroused concern all over the world (Hlihor and Gavrilescu, 2009; Ho and El-Khaiary, 2009; Gradinaru *et al.*, 2010). The demand for cleanup of contaminated environmental compartments to meet regulatory limits, as well as the increasing value of some metals place a call for efficient and low-cost treatment and metal recuperation technologies. Conventional methods for heavy metals removal from industrial effluents include precipitation, ion exchange, electrochemical methods and reverse osmosis (Kurniawan *et al.*, 2006; Popa ***et*** *al.*, 2010; Hong and Ning, 2011; Katsou *et al.*, 2011). Chromium(VI), one of Cr oxidation states is a suspected carcinogen and a potential soil, surface water, and groundwater contaminant. Although Cr(VI) may occur in the natural environment, human-caused Cr(VI) contamination has recently been the focus of much scientific discussion, regulatory and legal concerns. Chromium (Cr) is widely used in surface treatments, stainless steel and alloy production, pigments, leather processing, production of catalysts

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(Singh and Gadi, 2009; Dunea and Iordache, 2011; Cvijovic *et al.*, 2011). Although there exist already established physico-chemical methods and processes for chromium removal from the environment, their application is sometimes restricted, due to technological or economical constrains, biosorption can represent a sustainable alternative for metal removal and/or recovery, based on metal-sequestering properties of dead or living biomass. The major advantages of biosorption technology are its effectiveness in reducing the concentration of heavy metal ions to very low levels and the use of inexpensive biosorbent materials (Volesky, 2007; Gadd, 2009; Wang and Chen; 2009; Owlad *et al.*, 2009; Chojnacka, 2010; Figueiredo *et al.*, 2010a, Figueiredo *et al.*, 2010b; Gavrilescu, 2010; Kicsi *et al.,* 2010; Pilli *et al.,* 2010; Ramesh *et al.*, 2011; Zhang *et al.,* 2011).

Many biological materials can bind heavy metals, but only those with sufficiently high binding capacity and selectivity for heavy metals are suitable for use in a full-scale biosorption and bioaccumulation processes (Gavrilescu, 2004; Gadd, 2009; Wang and Chen; 2009; Karaduman *et al.,* 2011; Gomes *et al.*, 2011). Biosorption not only offers an innovative alternative to other remediation approaches, but it also allows metals recovery (Kotrba, 2011; Viraraghavan and Srinivasan, 2011). Among the different types of biomass proposed for heavy metals bioremediation, yeast cells of *Saccharomyces cerevisiae*, seems to be a promising alternative (Cojocaru *et al.*, 2009; Machado *et al.*, 2010a; Machado *et al.*, 2010b; Zhang *et al.*, 2011).

The aim of the present paper is a better understanding of Cr(VI) biosorption/reduction by heat inactivated yeast biomass (*Saccharomyces cerevisiae*), in a batch system by varying pH, biosorbent dosages, contact time and temperature, as well as to elucidate the mechanism of metal uptake by *S. cerevisiae* that represents a real challenge in the field of biosorption, thorough kinetic modeling of the bioremoval of heavy metal by *S. cerevisiae* and FTIR and SEM analysis.

MATERIALS & METHODS

Commercially distributed 'active' wet pressed biomass of *Saccharomyces cerevisiae* was supplied from a local store. Biomass was washed three times in distilled water followed by centrifugation at 6,000 rpm for 10 min. Yeast biomass was inactivated by heating in an oven at 80 °C for 24 h (Hlihor *et al.*, 2010). The dried yeast was grounded in a mortar to a constant particle size of 125-250 μm. In order to test the absence of viability of heat inactivated biomass an additionally procedure was applied: the dried cells were suspended in sterilized distilled water and incubated for one week at 28 ºC on agar plates. After this time period, no living colonies were found, so that it was demonstrated that all the yeast population was dead. All chemicals used in this work were of analytical reagent grade and were used without further purification. Chromium stock solution of 1000 mg/L was prepared by dissolving $K_2Cr_2O_7$ (Riedel) in distilled water. The solution was diluted for different metal concentrations by distilled water as required by the working procedure. Samples of Cr(VI) were analyzed spectrophotometrically by measuring the absorbance of the pink colored complex of Cr(VI) with 1,5-diphenylcarbazide in acidic solution at 540 nm (T60 UV-Visible Spectrophotometer) as described in the Standard Methods (Clesceri *et al.*, 2005). For total Cr concentration analysis, the Cr(III) was oxidized to Cr(VI) at high temperature by the addition of an excess of potassium permanganate prior to the 1,5-diphenylcarbazide reaction. The concentrations of Cr(III) were then obtained by the difference between total Cr and Cr(VI) concentrations (Clesceri *et al.*, 2005).

The biosorption of Cr(VI) was examined in batch in 150 mL conical flasks by mixing the desired amount of inactivated biomass with 50 mL solution of known Cr(VI) concentration and agitated at 150 rpm. A series of assays, with 100 mg/L Cr(VI) were conducted under different pHs (1-4) and room temperature (25±1°C) to investigate the influence of pH on Cr(VI) biosorption by inactivated *S. cerevisiae*. In the case of optimum biomass dosage determination, 1 to 8 g/L *S. cerevisiae* were used. For initial Cr(VI) concentration variation with time, concentrations of 25, 50, 100 and 200 mg/L were used, and the experiments were performed at different temperatures (25 ºC, 40 ºC and 50 ºC). For desorption experiment, the Cr-laden biomass was obtained through contact of 50 mg/L Cr(VI) pH 2 and 50 ºC for 24 h, long enough to reach 100% Cr(VI) reduction. Then the experiment was continued at 50 ºC with 50 mL H_2SO_4 and NaOH (0.5 M) as eluants (Park *et al.*, 2005). Samples were taken at predetermined time intervals for 30 days, and analyzed for Cr(VI) and total Cr. The initial pH of the working solutions was adjusted by addition of 0.1M H_2SO_4 and 0.1M NaOH solutions. The change in the working volume due to the addition of H_2SO_4 and NaOH was negligible. The pH measurements were performed with a HANNA precision pH meter, model pH 213. The batch experiments were carried out in an orbital incubator IKA KS 4000 IC control. All the experimental data were the averages of duplicate or triplicate experiments. All glassware used for experimental purposes was washed in 20% nitric acid and rinsed with distilled water to remove any possible interference by other metals.

Infrared spectra of heat inactivated *S. cerevisiae* biomass with and without the metal ion were acquired using JASCO FT/IR-4200 Spectrometer. The

powdered dried samples after the biosorption process were analyzed in the range 3500-600/cm with a resolution of 4/cm by using the ATR technique and placing enough sample to cover the diamond sensor.

SEM of the heat inactivated *S. cerevisiae* biomass with and without the metal ion was taken with a VEGA II LSH SEM (Czech Republic). The cell samples coated with carbon were examined under high vacuum mode.

Metal uptake in time and at equilibrium, and biosorption efficiency of heat inactivated biomass were determined according to Eqs. (1-3):

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

$$q_t = \frac{C_i - C_e}{m} V \quad (2)$$

$$R\,(\%) = \frac{C_i - C_e}{C_i} \times 100 \quad (3)$$

where: q_e and q_t are the amount of metal removed from solution (mg/g) at time t and at equilibrium; $R(\%)$ is the biosorption efficiency; C_i and C_e are the concentrations (mg/L) of metal ion in the initial solution and at the equilibrium after the experiment; V (L) is the volume of the solution; m (g) is the amount of biomass used in the experiment.

Desorption efficiency was given as (Eq. 4):

$$D\% = \frac{amount\ of\ metal\ ions\ desorbed}{amount\ of\ metal\ ions\ adsorbed} \times 100 \quad (4)$$

Different modeling approaches are used to represent the kinetic data of heavy metal biosorption. Distinctive adsorption kinetic models are of extensive use in explaining the biosorption mechanism of heavy metals (Gavrilescu, 2004; Pagnanelli, 2011). In spite of the importance of adsorption equilibrium studies, that determine the efficacy of adsorption, kinetic models have been exploited on the purpose of investigating the mechanism of biosorption and its potential rate-controlling steps. In addition, information of the kinetics of metal uptake is required to select the best conditions for full-scale batch metal removal process (Febrianto *et al.*, 2009).

In order to investigate the mechanism of sorption various kinetic models have been suggested and are generally expressed in linear form as follows:

Lagergren pseudo-first order model (Smaranda *et al.*, 2011)

$$\log(q_e - q_t) = \log q_e - \frac{k_1}{2.303} t \quad (10)$$

Pseudo-second order model (Ho, 2006)

$$\frac{t}{q_t} = \frac{1}{k_2 q_e^2} + \frac{1}{q_e} t \quad (11)$$

*Elovich model (*Gupta and Bhattacharya, 2011)

$$\frac{1}{q_t} = \frac{ln(\alpha\beta)}{\beta} + \frac{ln\, t}{\beta} q \quad (12)$$

where q_e and q_t (mg/g) are the adsorption capacity at equilibrium and at time t (min), respectively k_1 is the rate constant of the pseudo-first order equation (per min), k_2 is the rate constant of pseudo-second order adsorption (g/mg min), α is the Elovich initial adsorption rate (mg/g min) and β is the Elovich desorption constant (mg/g min) during one experiment.

Some authors have recently reported that the biosorption mechanism of Cr(VI) by biomaterials is not "anionic adsorption" but "adsorption-coupled reduction" (Park *et al.*, 2007), therefore kinetic models based on reduction were also applied to fit the experimental data of Cr(VI) bioremoval by *S. cerevisiae* (Eqs. 13, 14) (Wu *et al.*, 2010):

Pseudo-first order reduction:

$$\frac{dC}{dt} = -k_3 C, \quad \ln C = \ln C_0 - k_3 t \quad (13)$$

Pseudo-second order reduction:

$$\frac{dC}{dt} = -k_4 C^2, \quad \frac{1}{C} = k_4 t + \frac{1}{C_0} \quad (14)$$

where C_0 and C are Cr(VI) concentrations in solution (mg/L) at time 0 and t respectively, and k_3, k_4, are the apparent rate constants. The kinetic models parameters were evaluated by linear regression using ORIGIN Pro8 software.

RESULTS & DISCUSSION

Due to the acidic nature of chromium-containing aqueous real solutions from specific industries, the pH experiments were conducted in the pH range 1-4 with 100 mg/L Cr(VI) and at 25 °C. Fig. 1 shows the effect of initial pH on Cr removal at various solution pHs, ranging from 1 to 4; pH profiles were investigated with no pH adjustments during the experiment. A preliminary study (Hlihor *et al.*, 2011) showed that in 5 d, Cr(VI) was completely removed from the aqueous solution by 5 g/L yeast biomass only at pH 1. In the

case of pH 2, 3 and 4 only 68.49%, 26.24% and 11.05% Cr(VI), respectively were removed from aqueous solution during the experiment (Hlihor *et al.*, 2011). Further experiments were conducted in order to achieve 100% for all the pH values, as shown in literature for other biomaterials (Park *et al.*, 2007; Wu *et al.*, 2010). As expected, the removal rate was strongly pH dependent; the effectiveness of the reduction of Cr(VI) to Cr(III) increases with decreasing the pH from 4 to 1 (Fig. 1a), and total Cr concentration decreased with increasing the pH from 1 to 4 (Fig. 1b). Although Cr(VI) reached 100% removal at pH 1-2, total Cr reached 32-36 % removal at this pH values and Cr(III) which initially was not present appeared in the solution (Fig. 1b). Heat inactivated *S. cerevisiae* could remove and reduce 71.8 and 76.8% of Cr(VI) to Cr(III), and 62.5 and 69.6% of total Cr at pH 3 and 4, respectively after 90 d. The increase of solution pH from 1 to 4 increases the negative charge on the cell surface due to the deprotonation of the metal binding sites hence attracting Cr(III) ions resulting from the reduction of Cr(VI) (Silva *et al.*, 2009). At low pH, $HCrO_4^-$ species are dominant and enter in reactions, when Cr(VI) is reduced to Cr(III). Depending on the pH of the solution, Cr(VI) species may be in the forms (Baral *et al.*, 2006; Silva *et al.*, 2009): dichromate ($Cr_2O_7^{2-}$), hydrochromate ($HCrO_4^-$), chromate (CrO_4^{2-}). Also, Cr(III) species may take the following forms (Silva *et al.*, 2009): hydrated trivalent chromium, $Cr(H_2O)_6^{3+}$, Chromium hydroxide complexes, $Cr(OH)(H_2O)_5^{2+}$ or $Cr(OH)_2(H_2O)_4^+$. These forms also determine the behavior at various pH values and intervals (Deng and Bai, 2004; Quintelas *et al.*, 2009): as a result of repulsive electrostatic interactions, Cr(VI) anion species are poorly adsorbed by the negatively charged groups and can move freely; Cr(III) species, which carry positive electric charges could be adsorbed on the negatively charged groups. The reactions leading to reduction of Cr(VI) to Cr(III) can be summarized as follows (Silva *et al.*, 2009):

$$Cr_2O_7^{2-} + 14H^+ + 6e^- \leftrightarrow 2Cr^{3+} + 7H_2O \quad (18)$$

$$CrO_4^{2-} + 8H^+ + 6e^- \leftrightarrow Cr^{3+} + 4H_2O \quad (19)$$

$$HCrO_4^- + 7H^+ + 3e^- \leftrightarrow Cr^{3+} + 4H_2O \quad (20)$$

$$H_2CrO_4^- + 6H^+ + 3e^- \leftrightarrow Cr^{3+} + 4H_2O \quad (21)$$

Taking into consideration the reduction of Cr(VI) and total Cr removal by inactivated *S. cerevisiae*, for next studies, pH 2 was selected as optimum pH. Fig. 2a shows the time-dependent concentration of Cr(VI) at various biomass dosages (1 to 8 g/L heat inactivated *S. cerevisiae*). The removal rate of 100 mg/L Cr(VI) increased with increasing the biomass dosage. 100% reduction of Cr(VI) to Cr(III) was possible for 3, 5 and 8 g/L inactivated *S. cerevisiae* at a contact time of 90 d, 12 d and 25 d. At 8 g/L biomass dosage, the removal rate was slower than at 5 g/L; this could be explained by the fact that the competition of the ions for the available sites caused a decrease of the removal efficiency with increment in the biosorbent dosage (Deng *et al.*, 2009). After a complete removal/reduction of Cr(VI), 31.6, 36.01 and 49.03 % of total Cr was bound to the biomass (Fig 2b) at 3, 5 and 8 g/L, respectively. For 1 g/L, even if the reduction of Cr(VI) was possible

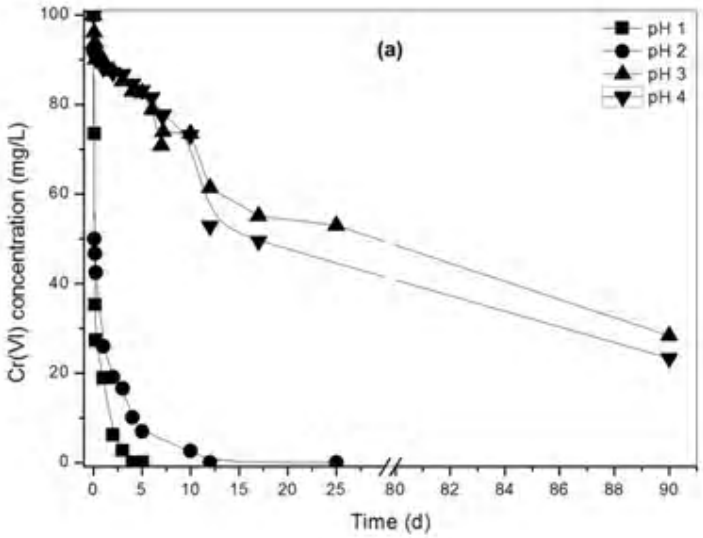
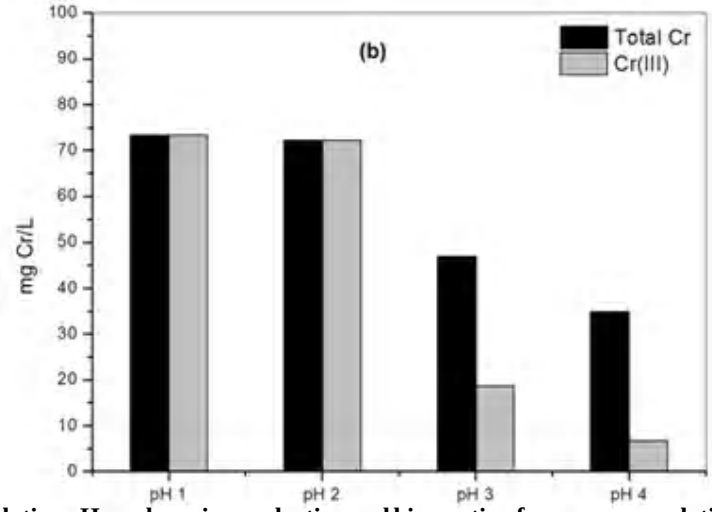


Fig. 1. Effect of solution pH on chromium reduction and biosorption from aqueous solution by heat inactivated *S. cerevisiae*: a) Cr(VI) (mg/L) vs. time, b) Total Cr (mg/L) and Cr(III) (mg/L) at the end of contact time (biomass dosage: 5 g/L; Cr(VI) concentration: 100 mg/L; temperature: 25°C)



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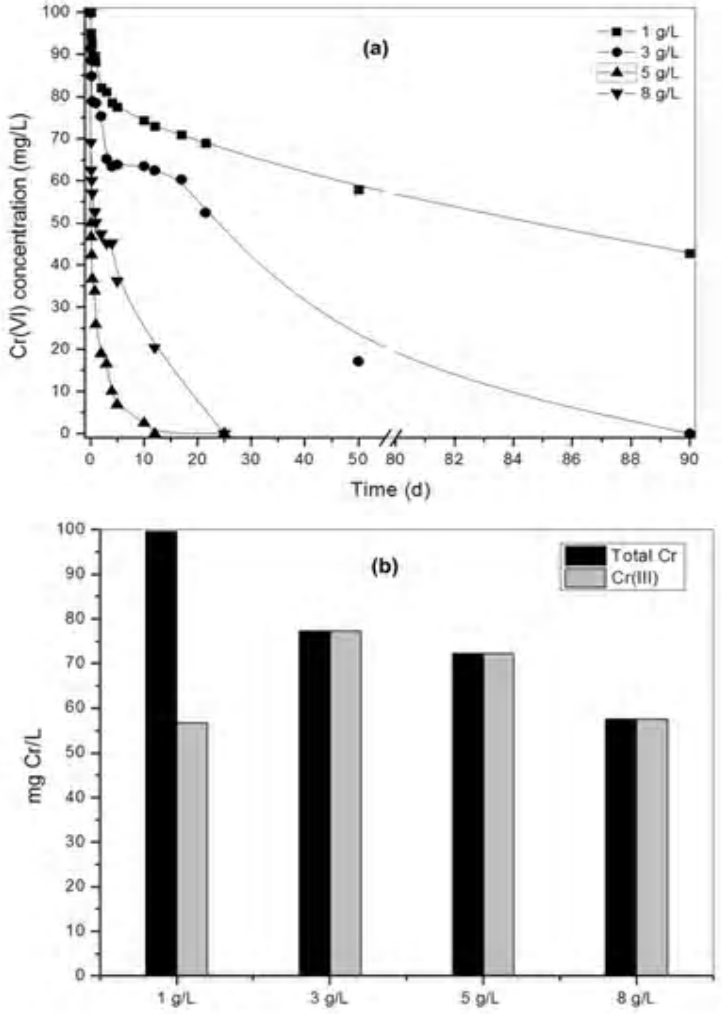

Fig. 2. Effect of biomass dosage on chromium reduction and biosorption from aqueous solution by heat inactivated *S. cerevisiae*: a) Cr(VI) (mg/L) vs. time, b) Total Cr (mg/L) and Cr(III) (mg/L) at the end of contact time (pH 2; Cr(VI) concentration: 100 mg/L; temperature: 25°C)

57.7%, the total Cr removal reached at the end of contact time (90 d), 11.82 %. The sorption on the surface is saturated faster at low biomass dosages. The availability of higher energy sites decreases with a larger fraction of lower energy sites occupied, resulting in a lower uptake value and higher removal efficiency at higher biomass dosages (Gupta and Rastogi, 2008). In further studies, pH 2 and 5 g/L inactivated *S. cerevisiae* were used for the reduction of Cr(VI) and total Cr removal from aqueous solution. The concentration of Cr(VI) versus time was examined at various initial Cr(VI) concentrations, from 25 to 200 mg/L and at different temperatures, 25, 40 and 50 °C; pH 2, 5g/L biomass dosage and 150 rpm were maintained constant. At 25 °C, the complete removal of 25 mg/L Cr(VI) required 8 d, while 200 mg/L Cr(VI) achieved a removal of 41 % in 17d (Fig. 3a). The increase of temperature greatly increased the Cr(VI) removal rate and decreased the contact time required for a complete removal of Cr(VI) (Fig. 3b, c). The necessary time for a complete removal of Cr(VI) at 50 °C was 2d and 17d for 25 and 200 mg/L, respectively. As mentioned in similar studies employing biomaterials, the increase of temperature induces the rate of the redox reaction (Park *et al.*, 2005). Cr(VI) could be completely removed from solution as long as the contact time was sufficient and with rise of the temperature; the mechanism involved in Cr(VI) removal by inactivated *S. cerevisiae* is therefore not ''anionic adsorption'' but ''adsorption coupled reduction'', as shown in previous works employing biomaterials (Park *et al.*, 2006; Deng *et al.*, 2009).

Because both adsorption and reduction could have an important impact in the removal of Cr(VI) from aqueous solution, the kinetic models based on adsorption and reduction were analyzed and the regressions parameters are reported in Table 1.It is indicated that for the experimental data regarding the influence of initial Cr(VI) concentration and temperature (25 and 50 ºC) (Fig. 4), the best fitting model is pseudo-first-order reduction model; in contrast, for the experiments performed at 40 ºC the best fitting model is pseudo-first-adsorption model.The experiments regarding the influence of pH and biomass dosage were best fitted by the pseudo-second-order reduction model. These results are indicating possible differences in the Cr(VI) removal mechanism at different experimental conditions. The values of R^2 indicated for the best fitting kinetic models are higher than 0.9. Based on the assumption that a biosorbent promotes an economic treatment of aqueous solutions, the possibility of metal recovery was studied through desorption experiments. Desorption of Cr bound to the biomass seems to be a difficult task considering the assumption that Cr(VI) was reduced to Cr(III). Several researchers noticed that solutions of alkali and highly alkaline salts (NaOH, Na_2CO_3 and $NaHCO_3$) were more potent than acids or mineral salts for Cr (VI) desorption (Isa *et al.*, 2008).

The desorption experiments were studied using 0.5M H_2SO_4 and 0.5M NaOH as eluants (Fig. 5a, b) at 50 °C, in batch. As seen in the fig, the 0.5 M NaOH is a more suitable eluant then H_2SO_4, but with a low desorption efficiency (12 %) of the Cr bound to the biomass in 30 d. 0.5 M H_2SO_4 eluted only the Cr(III), while both Cr(VI) and Cr(III) were eluted by 0.5M NaOH. In this case, Cr(III) appeared only until 150 min and disappeared from the solution slowly after 240 min. Based on this results, it is possible that both Cr(III) and Cr(VI) exist on the biomass and the latter might be reduced to the former which was eluted into the aqueous solution (Park *et al.*, 2005). This study suggests that a higher concentration of NaOH may be required to increase desorption of possible Cr species bound on *S. cerevisiae* biomass. A further study was not performed, since a more concentrated NaOH solution would raise the cost of a wastewater treatment, putting under discussion the suggested low-cost technology.

FTIR and SEM analysis were used as techniques for biosorbent characterization, as well as to explore the possible mechanism involved in the removal of Cr(VI) by *S. cerevisiae*. The FTIR spectrum of dried *S. cerevisiae* before and after biosorption of Cr(VI) is shown in Fig. 6. The unloaded yeast biomass, presents a broad band in the region 3750-3230/cm, with a maximum at 3277.4/cm; this peak is present due to hydroxyl (OH) stretching vibrations bonded with N-H stretching. Methylene C-H stretching bands typically assigned at 2940 – 2915/cm are present with a peak at 2924.5/cm. The peaks at 1638.2/cm and 1535.05/cm are the result of the stretching vibrations of C=O (carbonyl) and N-H(amide) deformation bands which result in IR bands between 1900-1550/cm and 1660-1500/cm, respectively. The band at 1396.2/cmresults in a bending vibration of COOH and OH groups (Dai *et al.*, 2008). The P=O stretching vibration occurs at 1040.4/cm. The phosphate out-of-phase stretch has strong IR bands between 1180 and 1000/cm.

After the retention of metal ion on the biomass, changes in intensity and shift in the peaks position occur. This indicates that heavy metal removal process by *S. cerevisiae* biomass doesn't imply the formation of stable covalent bonds, but is predominantly realized by chemical interactions (ion-exchange). Chromium ions have strong interactions with functional groups from peaks 1515.7/cm (C=O (carbonyl) and N-H(amide) deformation bands) and 1036.5/cm (P=O stretching). In these regions, there is an enhancement in intensity, due to chromium interaction.

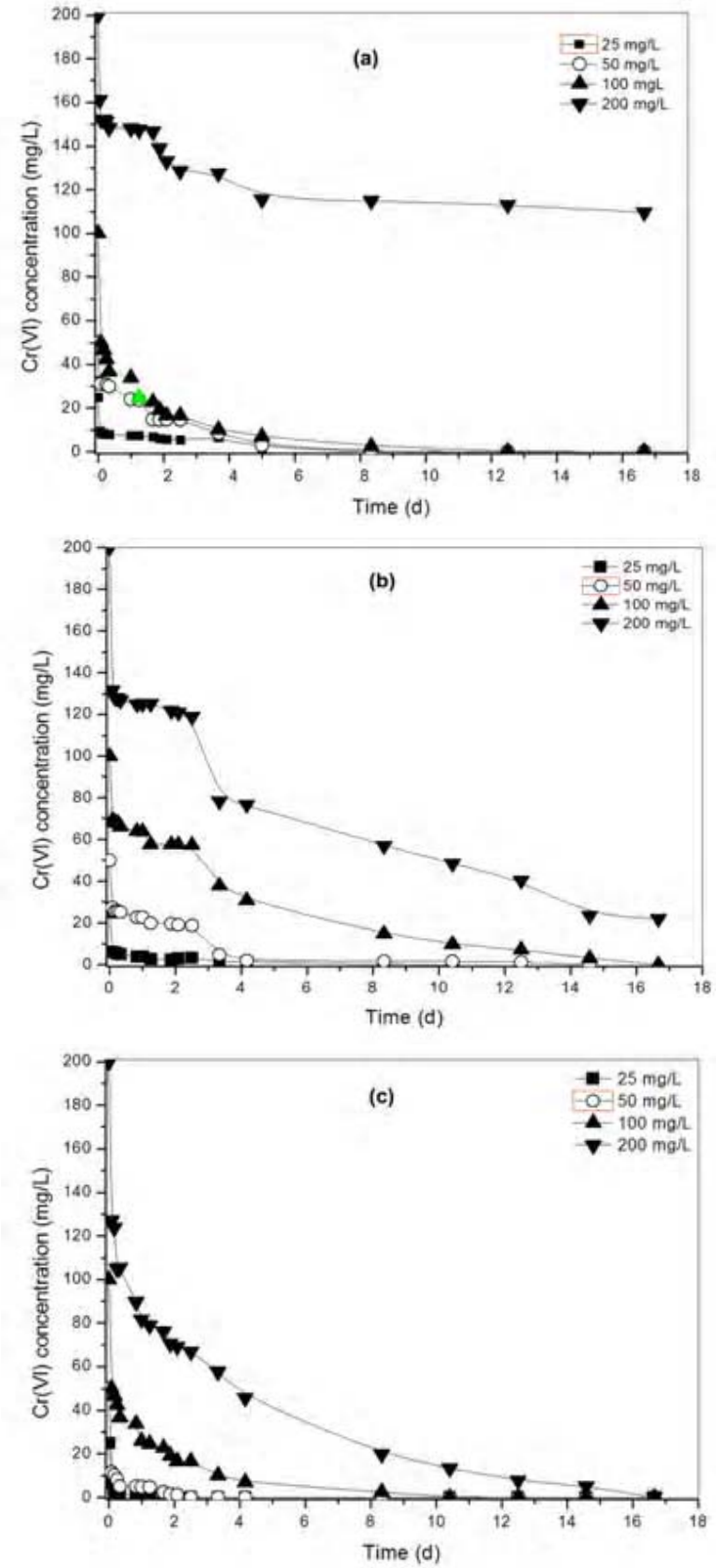

Fig. 3. Effect of contact time on Cr(VI) reduction by heat inactivated *S. cerevisiae* at different temperatures: a) 25 °C; b) 40 °C; c) 50 °C (pH 2; biomass dosage: 5 g/L)

Table 1. Kinetic parameters obtained for Cr(VI) removal by heat inactivated *S. cerevisiae*

Kinetic models	Item	Value	Pseudo-first order		Pseudo-second order	
			k	R^2	k	R^2
Reduction kinetics	Cr(VI) concentration (mg/L)25 °C	25	0.0125	0.9016	0.0027	0.5728
		50	0.0196	0.9559	0.0021	0.7120
		100	0.0148	0.9763	0.0014	0.9271
		200	0.0017	0.833	$7.27\ 10^{-6}$	0.7788
	Cr(VI) concentration (mg/L) 40 °C	25	0.0171	0.8456	0.0051	0.8901
		50	0.0125	0.7226	0.0021	0.8716
		100	0.0081	0.9541	$4.88\ 10^{-4}$	0.8608
		200	0.0038	0.9329	$5.85\ 10^{-5}$	0.9330
	Cr(VI) concentration (mg/L) 50 °C	25	0.0016	0.3225	0.0012	0.3259
		50	0.0418	0.901	0.0134	0.7768
		100	0.0153	0.9600	0.0017	0.9254
		200	0.0087	0.9948	$4.37\ 10^{-4}$	0.8682
	pH	1	0.0414	0.9339	0.0046	0.8876
		2	0.0148	0.9763	0.0014	0.9271
		3	$5.55\ 10^{-4}$	0.8871	$1.14\ 10^{-5}$	0.9769
		4	$6.40\ 10^{-4}$	0.8817	$1.50\ 10^{-5}$	0.9697
	biomass dosage (g/L)	1	$3.40\ 10^{-4}$	0.9089	$5.45\ 10^{-6}$	0.9599
		3	0.0012	0.9237	$3.39\ 10^{-5}$	0.9271
		5	0.0148	0.9763	0.0014	0.9271
		8	0.0126	0.9594	0.0014	0.9296
Adsorption kinetics	Cr(VI) concentration (mg/L)25 °C	25	0.0283	0.7624	0.0137	0.5728
		50	0.0147	0.9232	0.0102	0.8671
		100	0.0149	0.9754	$4.99\ 10^{-4}$	0.9493
		200	0.0075	0.8865	0.0011	0.9439
	Cr(VI) concentration (mg/L)40 °C	25	0.0138	0.9269	0.0258	0.8901
		50	0.0118	0.8011	0.0124	0.8372
		100	0.0070	0.9753	0.0024	0.8608
		200	0.0054	0.9360	$6.07\ 10^{-4}$	0.9719
	Cr(VI) concentration (mg/L)50 °C	25	0.0016	0.3225	0.0063	0.3259
		50	0.0418	0.901	0.0671	0.7769
		100	0.0153	0.9600	0.0089	0.9254
		200	0.0087	0.9948	0.0021	0.8682
	pH	1	0.0414	0.9339	0.0234	0.8876
		2	0.0149	0.9754	$4.99\ 10^{-4}$	0.9493
		3	0.0021	0.9586	$2.52\ 10^{-4}$	0.9186
		4	0.0023	0.8583	$2.73\ 10^{-4}$	0.7956
	biomass dosage (g/L)	1	0.0021	0.7574	$5.42\ 10^{-5}$	0.8140
		3	$6.05\ 10^{-5}$	0.6750	$3.70\ 10^{-5}$	0.7405
		5	0.0149	0.9754	$4.99\ 10^{-4}$	0.9493
		8	0.0037	0.9416	0.0046	0.9250

Fig. 7 presents the SEM micrographs of heat inactivated *S. cerevisiae* before (Fig. 7a) and after chromium binding (Fig. 7b). The inactivated biomass showed a porous structure and heterogeneous morphology, with pores with both larger and smaller dimensions. The yeast cells were oval and their surface appears to be smooth. The SEM micrograph of Cr-loaded yeast, indicates that the surface morphology doesn't change much; this leads to the conclusion that Cr(VI) reduction occurs at the interface of the adsorbent. FTIR and SEM analysis confirm that Cr(VI) can be directly reduced to Cr(III) in aqueous phase by contact with the electron-donor groups and that the release of the Cr(III) ions into the aqueous phase is due to electronic repulsion between the positively charged groups and the Cr(III) ions, or due to complexation of Cr(III) with adjacent groups capable of Cr-binding.

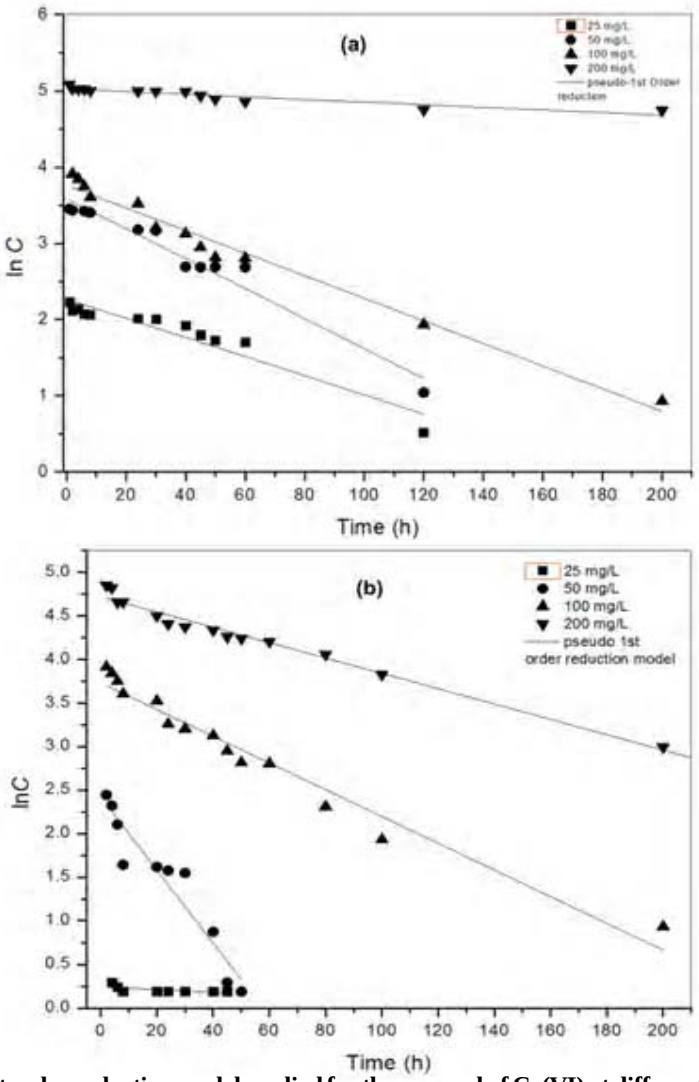

Fig. 4. Pseudo-first order reduction model applied for the removal of Cr(VI) at different temperatures by heat inactivated *S. cerevisiae*: a) 25 ºC b) 50 ºC

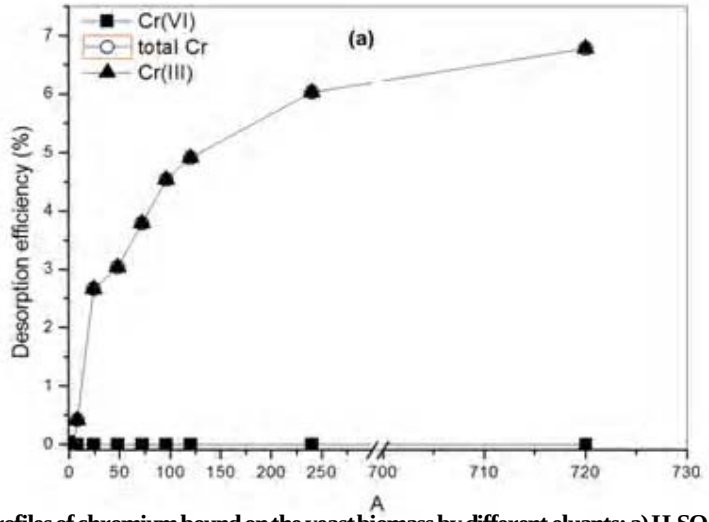

Fig. 5. Desorption profiles of chromium bound on the yeast biomass by different eluants: a) H_2SO_4 0.5M; b) NaOH 0.5M

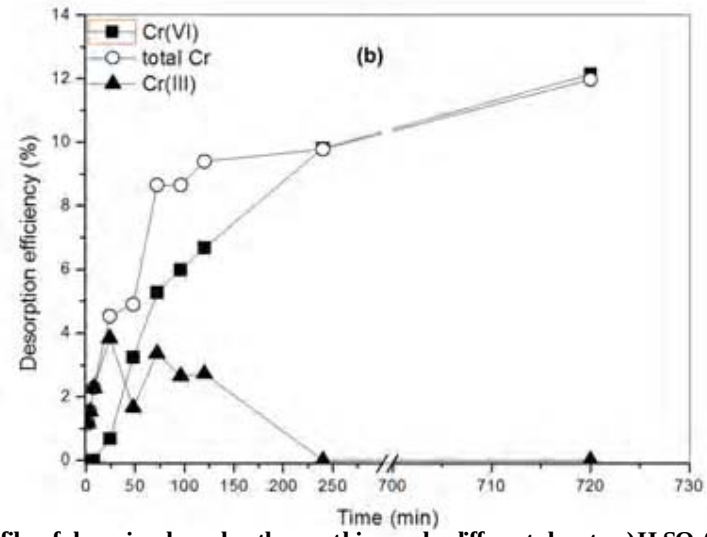

Fig. 5. Desorption profiles of chromium bound on the yeast biomass by different eluants: a) H_2SO_4 0.5M; b) NaOH 0.5M

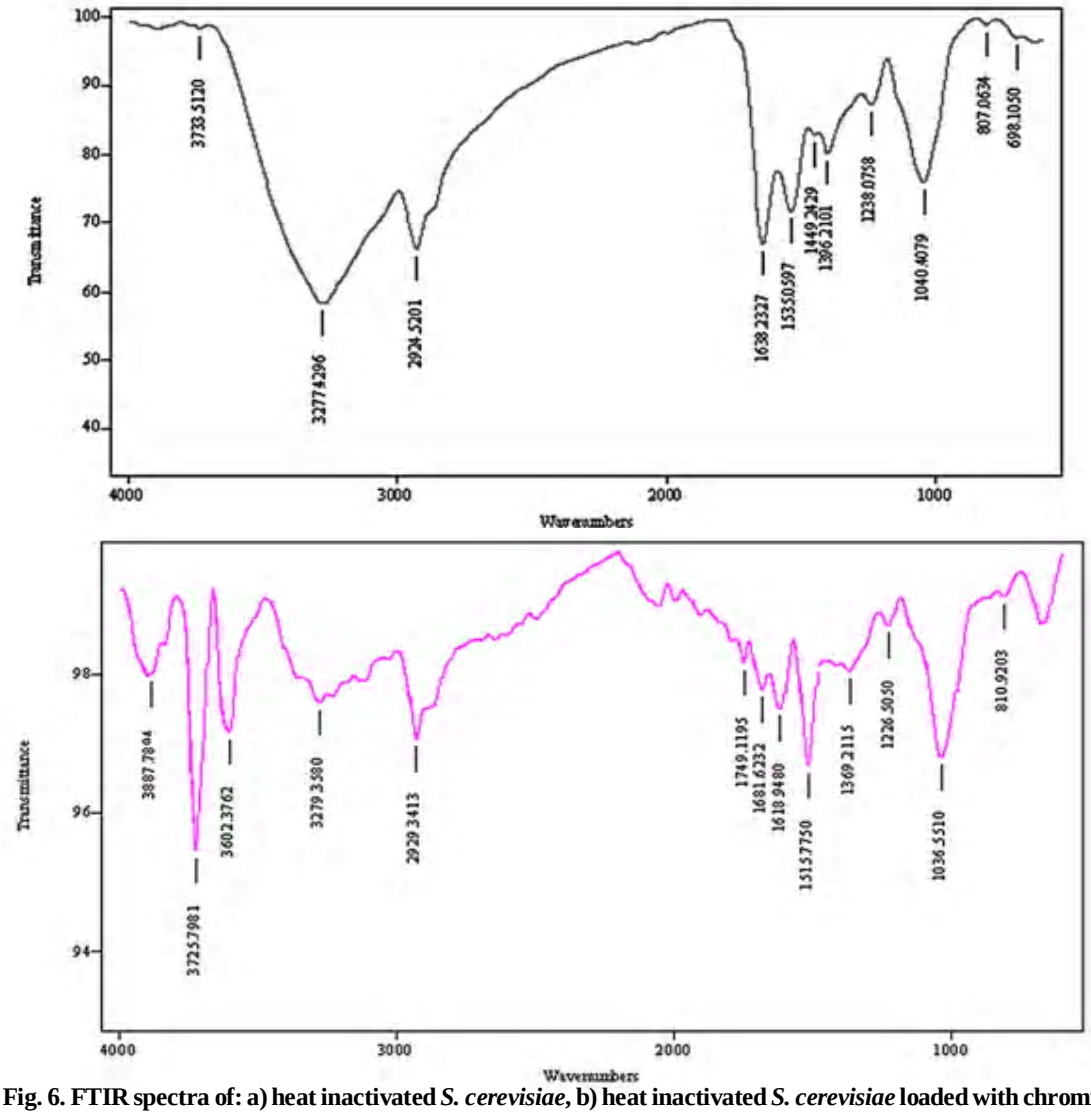

Fig. 6. FTIR spectra of: a) heat inactivated *S. cerevisiae*, b) heat inactivated *S. cerevisiae* loaded with chromium

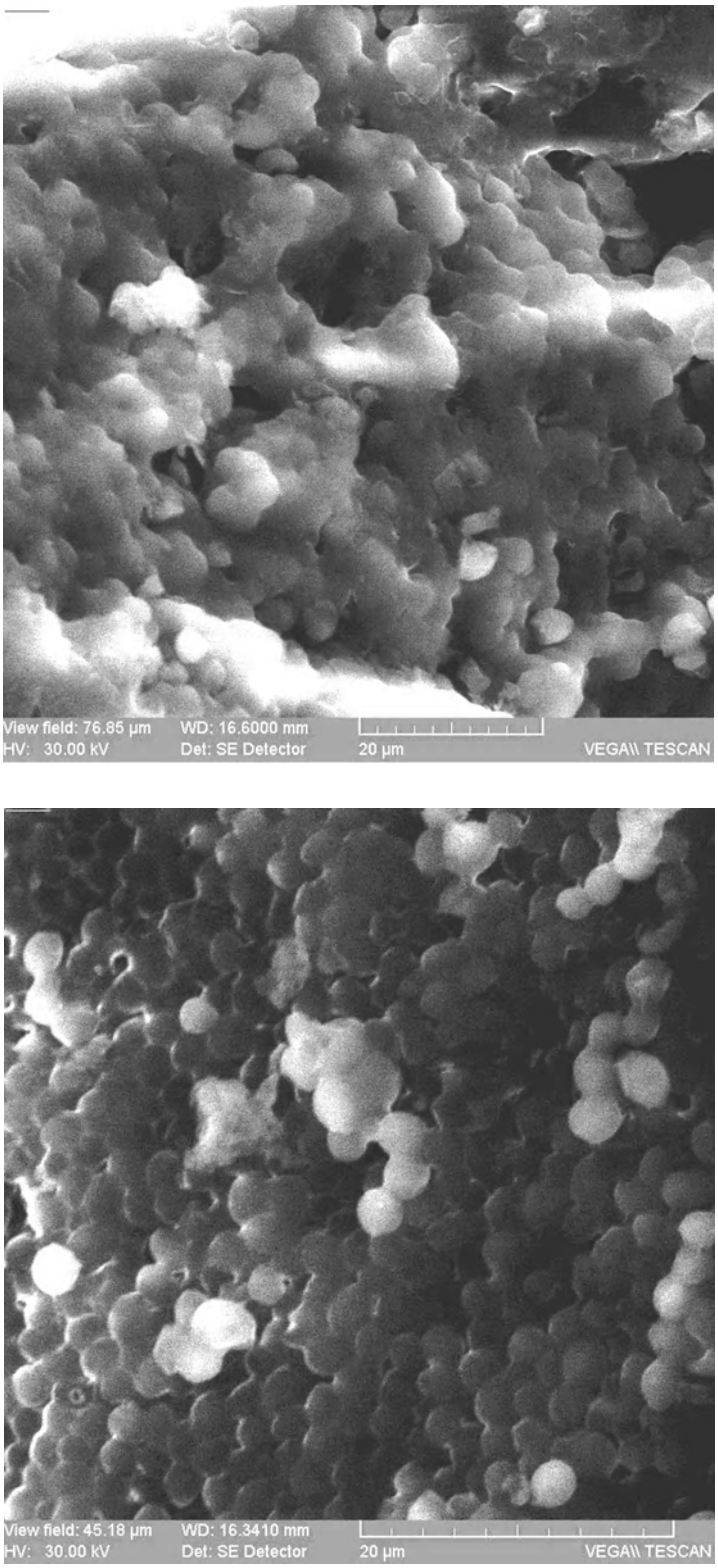

Fig. 7. SEM micrographs of: a) heat inactivated *S. cerevisiae,* b) heat inactivated *S. cerevisiae* loaded with chromium

CONCLUSIONS

The inexpensive *S. cerevisiae* yeast, produced in large quantities in the food and beverage industry could successfully remove Cr(VI) from aqueous solutions. Chromium reduction and biosorption was highly pH dependent, 100 mg/L of Cr(VI) being completely removed at pH 1-2 in 5-12 days by 5 g/L yeast biomass. Adsorption and reduction models were applied to the experimental data of the removal of Cr(VI) from aqueous solution. The experiments regarding the influence of pH and biomass dosage follow the pseudo-second-order reduction model while the experiments regarding the influence of initial Cr(VI) concentration and temperature (25 and 50 ºC) follow the pseudo-first-order reduction model. Results based on experimental data and FTIR and SEM analyses revealed that Cr(VI) can be directly reduced to Cr(III) in aqueous phase by contact with the electron-donor groups of the biomass release and the converted Cr(III) can be adsorbed to various functional groups of the biomass depending especially of the pH values. It can be concluded that the heat inactivated *S. cerevisiae* is an effective and alternative biomass for the removal of Cr(VI) form aqueous solutions, especially because it represents a by-product of fermentation industry, being produced in large quantities.

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