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Research article

Risk of low stability *Saccharomyces cerevisiae* ATCC 9763-heavy metals complex in gastrointestinal simulated conditions

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HIGHLIGHTS

- Biodecontamination of heavy metals from multi-metallic aqueous solutions using *Saccharomyces cerevisiae*.
- Stability assessment of metal-yeast complex after simulated gastrointestinal condition.
- Usingpretreatment strategies to increase bioremoval efficiency and stability.
- Indicating reversible bonds of heavy metal-yeast complexes.
- The Langmuir isotherm model was the best-predicting biosorption model.

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ABSTRACT

The biosorption of heavy metals by microorganisms has attracted the interest of food researchers as the last approach to reduce the risk of their absorption in the human body. But the stability of yeast-metal complexes under simulated gastrointestinal conditions has not been investigated. In this study stability of complex as well as isotherm and kinetic models of biosorption have been studied. Also, the impact of some pretreatment on yeast biosorption was studied to check the possible impact of different environmental conditions in food processing. Data showed a risk of heavy metal release in simulated gastrointestinal conditions. The best biosorption of metals from aqueous solutions by *Saccharomyces (S.) cerevisiae* may be achieved after NaOH pretreatment for Mercury (Hg) 92.7%. While biosorption of Lead (Pb) 37.48%, Arsenic (As) 19.44%, and Cadmium (Cd) 39.9% by untreated yeast were better. In gastrointestinal conditions, Hg and Cd-yeast complexes were more stable and biosorption of Cd and Pb increased. Bonds of As and Hg-yeast complexes in digestion conditions were reversible. The metals biosorption by untreated yeast followed the pseudo-second-order kinetic and the Langmuir isotherm model for Hg, Pb, and Cd and Freundlich for As. Results showed that biosorption of heavy metals by *S. cerevisiae*, although may decrease metal bioavailability in fermented foods, the complex is not enough stable in gastrointestinal conditions.

1. Introduction

Heavy metal contamination of water sources and the dangerous effects of these metals on human health are some of the most important scientific problems of the last decades (Raikwar et al., 2008). They can lead to kidney and liver cancer, reproductive disorders, disruptive effects on hematological and neurological systems, and cells' metabolisms (Mahurpawar, 2015). The acceptable dose of As, Hg, Pb, and Cd in drinking water are 10, 6, 10, and 3 μg L^{-1} to the World Health Organization (WHO) (Water and Organization, 2006).

There are various strategies for heavy metal removal, including physical and chemical methods as well as physicochemical and biological

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processes; in which, yeast, bacteria, seaweeds, and plants are used. Biosorbents of biosorption (biological adsorption) include fungi, bacteria, yeast, and plants (Malik et al., 2019). *Saccharomyces* (*S.*) *cerevisiae* is a precious biosorbent because of its wide use in fermented foods and drinks such as beverages, high safety, growth on inexpensive media, perfect model for kinetic and adsorption isotherm studies, and biomass production via simple fermentation methods (Savastru et al., 2019; Wang and Chen, 2006). Various studies have been carried out on bio removal using this yeast (Massoud et al., 2019). Results have verified the ability of *S. cerevisiae* in biosorption of all metals and heavy metals, even As. The bioremoval ability of yeast depends on various factors such as initial yeast biomass, initial heavy metal concentration, pH, ambient temperature, presence of other heavy metal ions, contact time, and composition of culture media (Hadiani et al. (2018a, 2018b); Hadiani et al., 2019). Naturally, *S. cerevisiae* includes phosphodiester bridges on its cell that produce negative surface charges. The mechanism of surface binding of *S. cerevisiae* is contributed to its surface charges (Zoghi et al., 2014). In addition, the groups of carboxyl, hydroxyl, amino, and phosphate in the cell wall of yeast are the principal responsible for heavy metal bioremoval (Fadel et al., 2017).

The content of heavy metals disposed into surface and groundwater is still increasing. Due to its dangerous potential to human life and the environment, there is a growing requirement for simultaneous monitoring of metals, such as Hg, Pb, As, and Cd in water samples (CONAMA, 2005; Mello et al., 2005). In previous reports of our research team, we studied the potential of yeast for biosorption of metals from food at low concentrations (ppb) from water and milk (Hadiani et al. (2018a, 2018b); Hadiani et al., 2019; Massoud et al., 2020, 2021). But there are a few studies on the ability of *S. cerevisiae* to remove low concentrations of Hg, Pb, As, and Cd in multi-metallic solutions and subsequent assessment of the bond stability of *S. cerevisiae* and heavy metals (simultaneous presence of these four metals in liquid phase) in simulated gastrointestinal conditions. In addition, for the first time, the comparison of various treatments for the uptake of heavy metals in ppb scales was examined. In this study, ethanol, caustic, and heat treatments were used to increase heavy metal biosorption. The current study aimed to assess the ability of *S. cerevisiae* for biosorption of As, Pb, Hg, and Cd in ppb scales and to study bond stability between these metals and *S. cerevisiae* under simulated gastrointestinal conditions. For finding the best bioremoval of As, Pb, Hg, and Cd, three treatments (caustic, ethanol, and heat) were compared, and for the best biosorption of these metals by yeast, three adsorption models (the Freundlich, Langmuir, and Temkin) were assessed using experimental data from the treated biomass. In addition, two kinetic models were used to anticipate the biosorption efficiency of these metals by the pretreated yeast biomass.

2. Materials and methods

2.1. Preparation of biomass and master culture and colony count

The *S. cerevisiae* ATCC 9763 was provided by Alzahra University Culture Collection in form of freeze-dried culture. The strain was cultured in nutrient broth [glucose, 1 g/50 ml distilled water (DW); yeast extract and NH_4Cl, 0.25 and 1 g/50 ml DW; KH_2PO_4 and Na_2HPo_4, 0.75 and 1.125 g/50 ml DW]. Yeast cultures were incubated at 27 °C for 16 h at 80 rpm (end of the exponential phase) and stored at 4 °C until use. For each series of bioremoval, seed cultures of *S. cerevisiae* were provided daily after inoculation of 5% (v/v) from the master culture. Then, seed cultures were agitated at 80 rpm for 16 h at 27 °C. The serial dilution method was used for the cell counting of seed cultures (Sieuwerts et al., 2008).

2.2. Chemical reagents

Components of the yeast cell cultures and the analytical reagents and chemicals were purchased from Merck, Darmstadt, Germany. Standard solutions of As (1000 mg/L in 0.1 M HNO_3) were provided by Panreac Quimica, Barcelona, Spain. Working standard solutions were prepared in deodorized water. Glass containers were soaked in 15% v/v HNO_3 for 24 h and then washed with deodorized water for the removal of elemental contamination. Then, glass containers were autoclaved for the removal of microbial contamination.

2.3. Saccharomyces cerevisiae pretreatments

For the heat pretreatment, yeast cells were sterilized at 121 °C for 20 min. Ethanol pretreated yeast cells were prepared by mixing yeast cells with 700 g/l ethanol solution at 25 °C for 1 h. The NaOH-treated cells were prepared by mixing yeast cells with 0.1 M NaOH and then incubating at 37 °C for 1 h. Treated yeast cells were centrifuged (4000 g, 10 min) and washed three times with deionized water. Pretreated yeast cells were used for As, Pb, Hg, and Cd biosorption (Göksungur et al., 2005).

2.4. Biosorption of heavy metals from aqueous solutions using Saccharomyces cerevisiae

Aqueous solutions were prepared by mixing 96.5 ml of sterile deionized water with 950 μL of As solution (10 ppm in 10% HCl), 525 μL of Cd and Pb (10 ppm in 0.1 M HNO_3), and 800 μL of Hg (10 ppm in 2% HCL) and adjusting the pH to 5 using 0.1 M NaOH and 0.1 M HCl. Concentrations of heavy metals were chosen based on the optimization of heavy metal biosorption by Hadiani et al. (2018a, 2018b) Final concentrations of the heavy metals in solutions included Cd and Pb, 52.5 μg/L; As, 95 μg/L; and Hg, 80 μg/L (Hadiani et al. (2018a, 2018b); Hadiani et al., 2019). Then, 1 ml of either untreated or pretreated *S. cerevisiae* solution (2.5×10^9 CFU/ml) was added to the solution and incubated at 25 °C for 24 h using a heater stirrer (Heidolph, German) (Hadiani et al., 2019). After 24 h, samples were added to gastric and small intestinal juices to estimate *the* stability of the *S. cerevisiae*-heavy metal (As, Pb, Hg, and Cd) bonds in gastrointestinal simulated conditions.

2.5. Preparation of gastric and small intestinal juices

For the preparation of simulated gastric juices, pepsin was added to a sterile NaCl solution (0.5%, w/v). The final concentration of pepsin was 3 g/L. Then, pH was adjusted to 2 using 30% HCl. Gastric juices were prepared daily and sterilized using 0.45-μm membrane filters (Nalge, Rochester, NY, USA). For the preparation of simulated small intestine juices; pancreatin (final concentration of 1 g. L^{-1}) and bile salts (final concentration of 1.5 g. L^{-1}) were added to sterile NaCl solution (0.5%, w/v). Then, pH was adjusted to 8.0 using 1 M NaOH. Small intestine juices were prepared daily and sterilized using 0.45-μm membrane filters (Nalge, Rochester, NY, USA) (Khorasani and Shojaosadati, 2017).

2.6. Selection of optimum biosorption conditions of Pb, As, Cd, and Hg by Saccharomyces cerevisiae ATCC 9763

In this study, the pH value was adjusted to 5. The initial concentration of metals in the solution was considered: Pb (52.5 μg/L), As (95 μg/L), Cd (52.5 μg/L), and Hg (79.8 μg/L), The temperature was selected at 25 °C, and initial concentration of yeast was considered approximately 10^7 CFU/mL Based on studies by Hadiani et al. (2018a, 2018b).

2.7. Bond stability between heavy metals and S. cerevisiae in gastrointestinal simulated condition

Briefly, 20 ml of each contaminated aqueous solution (25 °C, 24 h, 130 rpm for heavy metal biosorption by *S. cerevisiae*) were added to 80 mL of simulated gastric juices (37 °C) and vortexed (Vortex-Genie 2, Scientific Industries, Bohemia, NY, USA) for 10 s. It was incubated at 37 °C for 2 h. After sampling for heavy metal analysis, 100 mL of the simulated intestinal juice (37 °C) were added to the solution and

incubated at 37 °C for 2 h. The solution was agitated alternatively. Then, sampling for heavy metal analysis was repeated (Yin et al., 2018).

2.8. Analysis of heavy metals using inductively coupled plasma-mass spectroscopy

Inductively coupled plasma-mass spectroscopy (ICP-MS) (Agilent 7500, Agilent Technologies, USA) was used in this study. Plasma parameters, including nebulizing argon flow, radio frequency (RF) generator power, plasma gas flow rate, resonance RF frequency, and auxiliary gas flow rate, were 0.8 L/min, 1200W, 12.2L/min, 24MHz, and 0.8 L/min respectively. The limit of detection (LOD) of the analyzer for Pb, As, Cd, and Hg were 1.0, 1.0, 0.5, and 0.5 μg/L, respectively. and the limit of quantitation (LOQ) for the highlighted chemicals was 3.3, 3.3, 1.7, and 1.7 μg/L, respectively. Three replicate measurements for each sample were carried out.

2.9. Adsorption kinetic studies

Untreated *S. cerevisiae* cells (2.5×10^9 CFU/mL) were transferred into 100 mL of deionized water contaminated with Pb (52.5 μg/L), As (95 μg/L), Cd (52.5 μg/L), and Hg (79.8 μg/L) (pH 5.0) on a rotatory shaker. The metal ion concentrations of the sample were assessed at eight-time intervals to explain the adsorption kinetics of the *S. cerevisiae* cells. Pseudo-first and pseudo-second-order kinetic equations were assessed for heavy metal (Pb, As, Cd, and Hg) adsorption by untreated *C. cerevisiae* strains (Zoghi et al., 2020).

2.10. Isotherm model studies

Five samples with various numbers of untreated *S. cerevisiae* cells (2.5×10^9 CFU/mL) were contacted with five various initial concentrations of heavy metals (Pb, As, Hg and Cd) (pH 5) for 24 h. Langmuir, Freundlich, Temkin and isotherm models were used to study biosorption isotherms. Parameters of isotherm models were achieved similarly as previously described (Chen et al., 2015). Regression coefficient values (R^2) and the sum of error squares (ERRSQ) were calculated to describe the best isotherm, demonstrating biosorption of the heavy metals by untreated *S. cerevisiae*. All experiments were carried out in triplicates.

2.11. Statistical analysis

All experiments were carried out in triplicates and data were shown as mean ± standard deviation (X ±SD). Data processing was carried out using Statistical Package for the Social Sciences (SPSS) Software v.22.0 (SPSS Institute, Chicago, IL, USA). One-way Analysis of Variance (ANOVA) was used to estimate *p*-values and confidence levels. In general, *p*-values less than 0.05 were considered significant.

3. Results and discussion

3.1. Effects of pretreatments (heat, NaOH, and ethanol) of S. cerevisiae ATCC 9763 on the removal of Pb, As, Cd, and Hg after 24 h of exposure

Scanning electron microscopy and Fourier transform infrared spectrometry techniques show that biosorption of metal ions is physical and occurs at the surface of the yeast (Zinicovscaia et al., 2020). For examining the effects of pretreatments on this surface adsorption different treatments were examined. Previous reports showed effects (positive or negative) of pretreatments on toxins (Zoghi et al., 2020) and heavy metal biosorption (Göksungur et al., 2005). In this study, the major aim was to find the best condition (with or without treatment) for decontamination of Hg, Pb, As, and Cd in multi-metal aqueous solutions by *S. cerevisiae*. Results showed that the best biosorption achieved at 25 °C, pH 5, 24 h of exposure, and inoculum size of 2.5×10^9 CFU/mL accompanied by NaOH pretreatment for Hg (92.7%) and untreated yeast for Pb (37.48%), AS (19.44%) and Cd (39.9%). All in all, in the present study untreated cell yeast, were abler to biosorption of simultaneous presence of Cd, As, and Pb compared to pretreated cells; but Hg had better uptake with NaOH pretreated cells. Higher bioremoval rates of Hg by NaOH-treated yeast cells compared to untreated ones were similar to a previous study by Göksungur et al. (2005). Cell walls of yeast play important roles in biosorption and treatment by NaOH (caustic treatment) might degrade cell walls of yeast by removing their protein groups. Consequently, the destruction of the cell membranes of yeast exposes intracellular components and enhances the efficiency of biosorption by creating further surface binding sites and causing further easier bonds between the yeast cells and the metal ions (Göksungur et al., 2005; Khosravi-Darani et al., 2020; Wang and Chen, 2006).

In contrast, in the study by Ghorbani et al. (2008) ethanol pretreated yeasts were able to bioremoval of Cd two times greater than untreated cell yeast. That reason might be described by the addition in the accessibility of heavy metals to the binding sites on the surface of yeast. In addition, following the study by Ghorbani et al. (2008) in the present study bioremoval of Hg by ethanol-treated cell yeast (92.7%) was greater than untreated cell yeast (84.33%). This can be explained by increasing the accessibility of the binding sites of yeast for bioremoval of Hg (Ghorbani et al., 2008). The mechanism of heat treatment for the enhancement of biosorption is like ethanol treatment by increasing the disposal of additional functional groups implicated in the bioremoval of metals (Soares and Soares, 2012).

According to the existence of these four metals simultaneously in an aqueous solution, all in all, untreated cells of yeast show the best potential for uptake of these metals at the same time, and usage of pretreatments in the uptake of these four metals at the same time is not advised. This can be explained by the selective and sometimes competitive abilities of *S. cerevisiae* in bioremoval of the simultaneous presence of Hg, Pb, As, and Cd in aqueous solutions.

Table 1 shows heavy metal uptake by untreated and treated cell yeasts and compares the potential of treated and untreated cell yeast in bioremoval of these metals.

3.2. Stability assessment of yeast-metal complexes under simulated gastrointestinal conditions

Bond stability of biosorbent and absorbed in gastrointestinal condition has an important role in examining the efficiency of biosorption. In recent years, there are several studies about the assessment of bond stability between biosorbent and absorbed (Ribeiro et al., 2021; Zoghi et al., 2020).

Liquid-phase concentrations of Hg, Pb, As, and Cd after 24 h of exposure to untreated, heat, NaOH, and ethanol pretreated *S. cerevisiae* ATCC 9763 are demonstrated in Figure 1 a (Hg), b (Pb), c (As), and d (Cd). All samples (except As) showed strong biosorption of heavy metals within 24 h. Samples exposed to simulated gastrointestinal conditions for 24 h are illustrated in Figure 1. Also, the stability of the metal-yeast complexes to these conditions was assessed. NaOH pretreated yeast cells showed the best ability of Hg biosorption (Figure 1a). After exposure to simulated stomach conditions, low levels of Hg were released from the culture of untreated yeasts. In complexes of pretreated yeast-metal after exposure to simulated stomach conditions, large quantities of Pb were released from the complex of yeast-Pb (Figure 1b). In NaOH and ethanol pretreated yeasts, bonds between As and yeasts could be released (Figure 1c). Furthermore, Cd in untreated and treated yeast cells could be released and bonds between Cd and yeasts could be released under simulated stomach conditions (Figure 1d). Therefore, it could be concluded that the adsorption of these metals was reversible. A similar result was reported by Bao Le (Le and Yang, 2019). for Cd bioaccessibility of Cd- *Pediococcus pentosaceus* suspension after simulated gastrointestinal conditions. Cd bioaccessibility after the simulated gastrointestinal condition was 44.7–46.8 %. According to this study, the bonds between Cd and *Pediococcus pentosaceus* were reversible.

The current results showed that *S. cerevisiae* included the best ability of Hg bioremoval, compared to other heavy metals. However, bonds of this complex (Hg-yeast) were reversible under simulated gastrointestinal conditions. Moreover, As had the lowest biosorption rate and the yeast did not have a good ability of As bioremoval. Under this condition and the simultaneous presence of these four metals in the liquid phase, biosorption of Pb and Cd increased under simulated gastrointestinal conditions. According to this study, physical and chemical adsorptions of metals by *S. cerevisiae* might occur simultaneously. Furthermore, binding reversibility suggested non-covalent electrostatic bonds (e.g., Van der Waals and hydrogen bonds).

According to our results, untreated cell yeasts showed more potential for biosorption of Hg, Pb, As, and Cd in a multi-metallic aqueous solution. So, untreated cell yeasts were selected for more studies.

3.3. Kinetic model studies

Heavy metal biosorption by *S. cerevisiae* naturally depends on the exposure time. The metal kinetic model is one of the most important tools for the assessment of sorption mechanisms. Kinetic models describe the quantity and mechanism of adsorbed Hg, Pb, As, and Cd by *S. cerevisiae* cell layers at various times (Tuzen et al., 2020). The pseudo-first-order equation is shown as Eq. (1) (Al-Hazmi, 2010):

$$\ln(q_e - q_t) = lnq_e - K_1 t \quad \text{Eq. (1)}$$

Where, K_1 is the first-order kinetic rate constant, q_e (μg/L): Amounts of adsorbed metals at equilibrium times (h), and q_t (μg/L): Amounts of the adsorbed metals at a given time (t). The linear pseudo-second-order equation is shown as Eq. (2) (Anene et al., 2016):

$$\frac{t}{q_t} = \frac{1}{K_2 q_e^2} + \frac{t}{q_e} \quad \text{Eq. (2)}$$

Where, K_2 is the second-order kinetic rate constant, q_e (μg/L): Amounts of the adsorbed metals at equilibrium times (h) and q_t (μg/L): Amounts of adsorbed metals at a given time (t). Time profiles of Pb, As, Hg, and Cd adsorption by untreated *S. cerevisiae* ATCC 9763 in aqueous solutions are shown in Figure 2a. Figure 2b illustrates R2 (correlation coefficient values) from the linear regression analysis. Kinetic model studies were compared to pseudo-first-order (not shown) and pseudo-second-order (Figure 2b). The Pb, Hg, As, and Cd biosorption kinetic models were fitted to pseudo-second-order and their correlation coefficients for pseudo-second-order were 0.9775, 0.9991, 0.9422, and 0.9897, respectively. Similar results were reported by Ghorbani et al. (2008) for bioremoval of Cd by *S. cerevisiae* that was controlled by pseudo-second-order kinetic mechanisms. In another study, the pseudo-second-order kinetic mechanism was best described by the bioremoval of lead by *S. cerevisiae* (Ghaedi et al., 2010). In addition, the best fit of the kinetics of biosorption of Cd, Pb, and, Cu by *S. cerevisiae* was the pseudo-second-order (Dutta et al., 2016). About biosorption of As by *S. cerevisiae*, the best kinetic mechanism was reported pseudo-second-order equation (Wu et al., 2012). Kinetic models play important roles in evaluating the initial qualities and efficiency of adsorbents, the time required for biosorption of metals, and the identification of the kind of mechanisms involved in bioremediation system (Febrianto et al., 2009; Kumar and Sivanesan, 2006; Savić and Vasi ć, 2006). Furthermore, Kinetic studies are done to obtain information on the nature of processes occurring in biosorption of metals by yeast. This fitted biosorption to pseud-second order suggested that biosorption of heavy metals by yeast is through physico-chemical interactions. Also, this kinetic model is based on chemical biosorption (Chwastowski and Staroń, 2022). The pseudo-second kinetic model revealed chemical biosorption mechanisms (Araújo et al., 2013).

Table 1. Hg, Pb, As, and Cd uptake% by untreated and pretreated yeast.

% Metal uptake	Untreated	Ethanol treated	Heat-treated	NaOH treated
Hg	84.33^{Db}	90.60^{Dc}	67.41^{Da}	92.70^{Cd}
Pb	37.48^{Bd}	7.20^{Ba}	29.65^{Cc}	11.58^{Bb}
AS	19.44^{Ad}	0.90^{Aa}	3.90^{Ac}	1.83^{Ab}
Cd	39.90^{Cd}	18.80^{Ca}	26.90^{Bc}	19.30^{Cb}

The initial concentration of Hg, Pb, As and Cd were 77.94, 52.03, 87.20, and 48.01 μg/L, respectively. Results are mean values of triplicate determinations and small and capital letters show statistical differences for data in rows and columns, respectively ($p < 0.05$).

3.4. Biosorption isotherms of the heavy metals

The equilibrium data might be used to design appropriate adsorption systems for large-scale uses. In equilibrium conditions (approximate untreated yeast cells of 10^7 CFU/mL, pH 5, 24 h of exposure, 25 °C) at various initial concentrations of Pb, As, Hg, and Cd, particular associations between liquid and solid phases could be qualified generally using three various isotherm models of Langmuir, Freundlich, and Temkin (Freundlich, 1906; Langmuir, 1918; Tempkin and Pyzhev, 1940). In heavy metal biosorption studies, characteristics of *S. cerevisiae*, including structure, functional groups, and surface area, included important roles in metal biosorption (Ertugay and Bayhan, 2010). The Langmuir form could be described using Eq. (3):

$$Q_e = Q_{max}[K_L C_e / (1 + K_L C_e] \quad \text{Eq. (3)}$$

Where Q_e (μg/mg) is the amount of metals in adsorbing equilibrium, C_e (μg/L) is the equilibrium concentration of the metals in aqueous solutions, Q_{max} (μg/mg) is the maximum amount of the adsorbed metals at high C_e (μg/L) and K_L (L/μg) is the Langmuir adsorption constant. The Freundlich form could be reported via the following Eq. (4):

$$Q_e = K_F \times C_e^{1/n_F} \quad \text{Eq. (4)}$$

Where K_F is the Freundlich constant, n_F is the experimental parameter (relating to bioremediation intensity), C_e (μg/L) is the equilibrium concentration of metals in aqueous solutions, and Q_e (μg/L) is the amount of metals in adsorbing equilibrium. The Temkin form could be described using Eq. (5):

$$Q_e = \beta\, ln\alpha + \beta\, lnC_e \ (\text{E.1}) \ ; \ \beta = RT/K_T \quad \text{Eq.(5)}$$

Where, K_T (J/mol) is the Temkin constant, α (L/g) is another Temkin constant (R = 8.314 J/mol.K), T (K) is the absolute temperature, C_e (μg/L) is the equilibrium concentration of metals in aqueous solutions and Q_e (μg/L) is the amount of metals in adsorbing equilibrium. Hg, Pb, As, and Cd biosorption isotherms are illustrated in Figure 3 a, b, and c. Three adsorption isotherms of Langmuir (a), Freundlich (b) and Temkin (c) were used in data. Steady adsorption equilibrium was created when concentrations of the adsorbed heavy metals (Q_e) were similar to the concentrations of desorbed metals and equilibrium value (C_e) was fixed. The equilibrium adsorption isotherms included important roles in designing adsorption processes. Figure 3 shows a regression analysis of the isotherms. Coefficients of correlation for Pb were calculated as 0.9779, 0.881, and 0.8965 respectively belonging to Langmuir, Freundlich, and Temkin isotherms. In the regression analysis of isotherms, coefficients of correlation for Hg were reported as 0.9719, 0.8796, and 0.9247 associated with Langmuir, Freundlich, and Temkin isotherms, respectively. The coefficients of correlation of As were 0.9388, 0.9618, and 0.8034 linked to Langmuir, Freundlich, and Temkin isotherms, respectively. In the regression analysis of isotherms, coefficients of correlation for Cd were 0.941, 0.8671, and 0.8387, which were associated with Langmuir, Freundlich, and Temkin isotherms, respectively. The bioremediation process of Hg, Pb, and Cd further matched with the Langmuir isotherm model. same results were reported by Massoud et al.

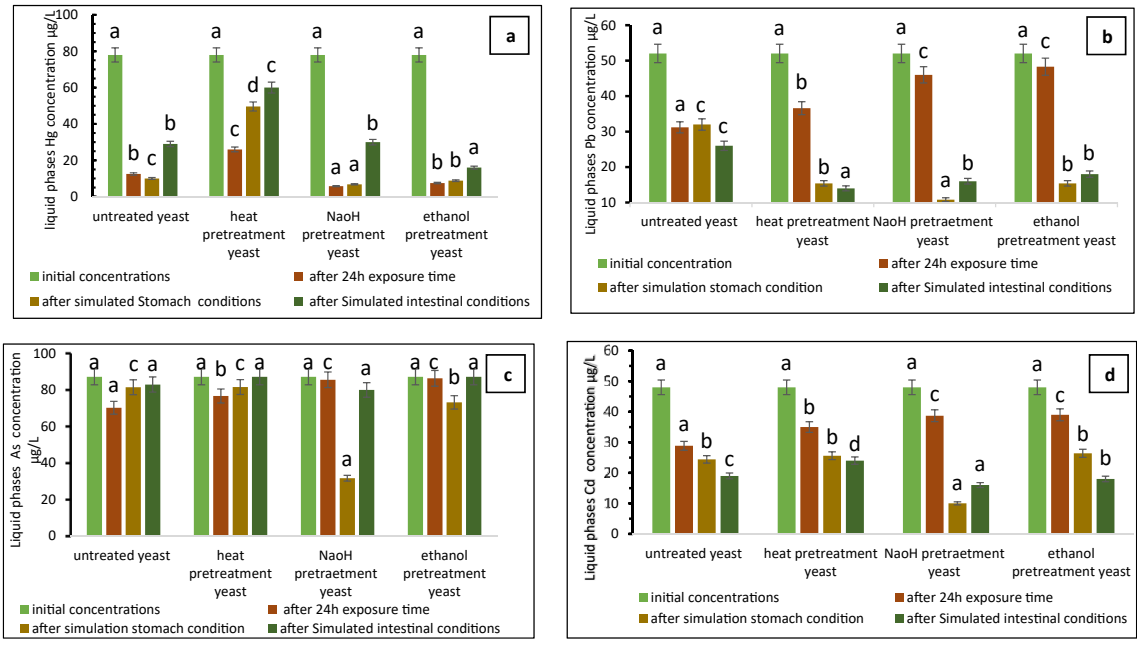

Figure 1. Heavy metals concentration (a = Hg, b = Pb, c = As, d = Cd) in Liquid phases after exposure to *S. Cerevisiae* ATCC 9763 after 24 h exposure time and Gastrointestinal conditions (comparison of untreated, and pretreated by heat, NaOH, and ethanol). Standard deviation is considered 95% confidence. small capital letters show statistical differences for data in the same columns ($p < 0.05$).

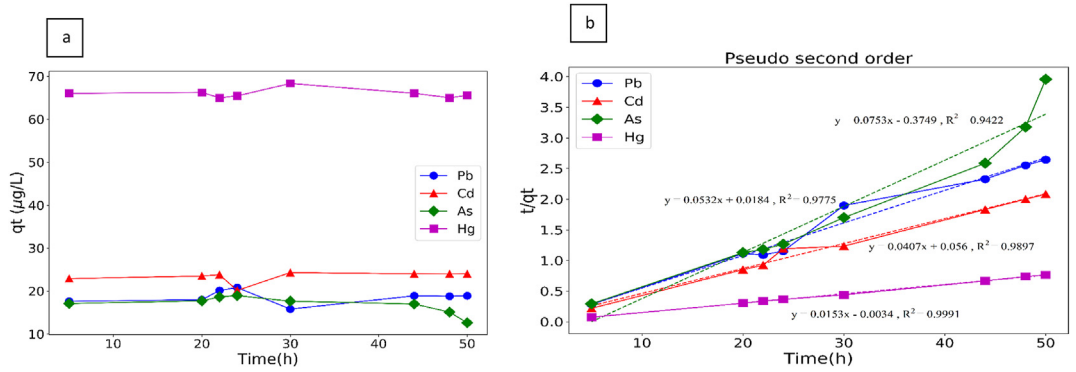

Figure 2. (a) Time profile for heavy metals adsorption. (b) Plot of the pseudo-second-order kinetic model of heavy metals adsorption by untreated *S. Cerevisiae* ATCC 9763 in aqueous solution. qt (µg/mg) and qe (µg/mg) were quantities of the adsorbed Heavy metals per milligram yeast cells at a given time (t) and equilibrium time (h), respectively.

(2020, 2021) for biosorption of Hg and Cd by *S. cerevisiae* were fitted to the Langmuir model (Massoud et al., 2020, 2021). Moreover, As matched with Freundlich model. The Langmuir model shows that the biosorption process was homogeneous, uniform and monolayer. In this isotherm model, suppressing chemical interactions were detected between the adsorbing molecules. In contrast, the Freundlich model shows that biosorption process was heterogeneous, non-uniform, and multi-layer and was not ideal (Chen et al., 2019). In the Freundlich isotherm model, chemical interactions were seen between the adsorbed molecules (Raoov et al., 2013).

4. Conclusions

In this study, the biosorption of heavy metals (pH 5, 24 h exposure time, yeast concentration approx. 10^7 CFU/mL and concentration of Cd and Pb, 52.5 µg/L; As, 95 µg/L; and Hg, 80 µg/L) by treated and untreated yeast cells was assessed. The results showed that the best-performing yeast for Pb, Cd, and As bioremoval was untreated and for Hg bioremoval was NaOH-treated cell yeasts. Furthermore, the yeast-metal complex under the simulated gastrointestinal condition was reversible. It can be concluded that metal binding to *S. cerevisiae* significantly depends on the metal and yeast cell wall structure. Untreated cell yeast had the best ability for biosorption of metals in multi-metallic aqueous solution. So, it was selected for the examination of kinetic and isotherms studies. Pb, As, Hg and Cd biosorption processes of untreated yeast cells followed the pseudo-second-order kinetic model. Among Langmuir, Freundlich, and Temkin adsorption isotherm models, the first shows high performance to predict the bio-removal efficiency of Pb, Cd, and Hg by untreated yeast cells. Freundlich isotherm model would be reliable to offer precise predictions of As biosorption.

Before this study, biosorption of metals and toxins by yeast has been considered a safe method in different fermented foods such as wine and fermented drinks (dough, kefir, kumis, etc). The data of this research showed the reversibility of metal-yeast bonds in gastrointestinal conditions. Such observation indicates that biosorption by yeast cannot be considered a safe approach for heavy metal bioremoval from food.

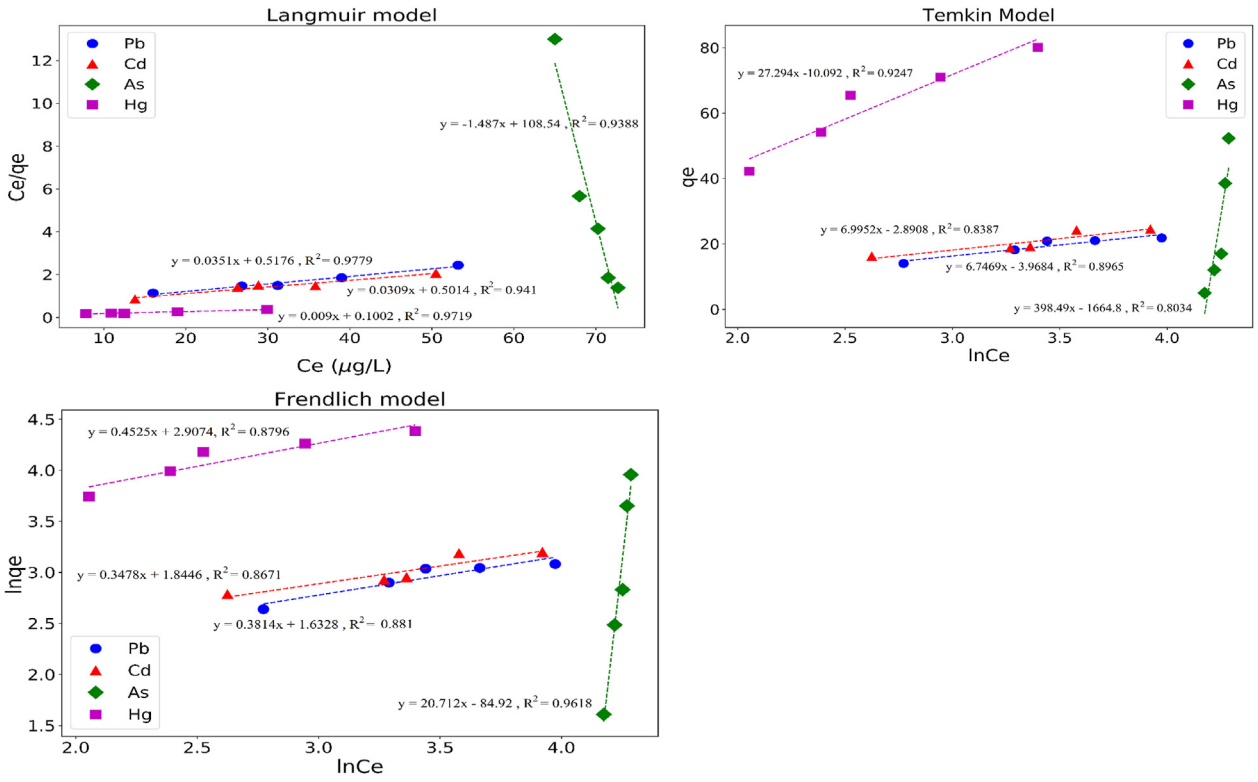

Figure 3. Plots for Langmuir, Freundlich, and Temkin adsorption isotherm curves of heavy metals adsorption by untreated *S. Cerevisiae* ATCC 9763 cells in contaminated aqueous solution are shown. Qe (μg/mg): quantity of heavy metals per mg yeast cells in adsorbing equilibrium, Ce (μg/L): equilibrium concentration of heavy metals in aqueous solution).

Further studies are needed to investigate the stability of the metal-yeast complex in various beverages and examine the reason reversibility and irreversibility of complex binding.

Declarations

Author Contribution statement

Razieh Sadat Mirmahdi: Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Vahid Mofid, Amir Mohammad Mortazavian: Contributed reagents, materials, analysis tools or data.

Alaleh Zoghi: Conceived and designed the experiments; Wrote the paper.

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The authors declare no conflict of interest.

Additional information

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References

Al-Hazmi, N., 2010. Determination of patulin and ochratoxin A using HPLC in apple juice samples in Saudi Arabia. Saudi J. Biol. Sci. 17 (4), 353–359.

Anene, A., Kalfat, R., Chevalier, Y., Hbaieb, S., 2016. Molecularly imprinted polymer-based materials as thin films on silica supports for efficient adsorption of Patulin. Colloids Surf. A Physicochem. Eng. Asp. 497, 293–303.

Araújo, C.S., Carvalho, D.C., Rezende, H.C., Almeida, I.L., Coelho, L.M., Coelho, N.M., Marques, T.L., Alves, V.N., 2013. Bioremediation of waters contaminated with heavy metals using Moringa oleifera seeds as biosorbent. Appl. Bioremed. Active Passive Approach. 23, 227–255.

Chwastowski, J., Staroń, P., 2022. Influence of *Saccharomyces cerevisiae* yeast cells immobilized on Cocos nucifera fibers for the adsorption of Pb (II) ions. Colloids Surf. A Physicochem. Eng. Asp. 632, 127735.

Chen, J., Dong, J., Shen, S., Mei, J., Chang, J., 2019. Isolation of the Hg (II)-volatilizing *Bacillus* sp. strain DC-B2 and its potential to remediate Hg (II)-contaminated soils. J. Chem. Technol. Biotechnol. 94 (5), 1433–1440.

Chen, Y., Zhang, B.-C., Sun, Y.-H., Zhang, J.-G., Sun, H.-J., Wei, Z.-J., 2015. Physicochemical properties and adsorption of cholesterol by okra (Abelmoschus esculentus) powder. Food Funct. 6 (12), 3728–3736.

CONAMA, 2005. National Council of Environment. *Resolution number 357, March 17*.

Dutta, A., Zhou, L., Castillo-Araiza, C.O., De Herdt, E., 2016. Cadmium (II), lead (II), and copper (II) biosorption on Baker's yeast (*Saccharomyces cerevesiae*). J. Environ. Eng. 142 (9), C6015002.

Ertugay, N., Bayhan, Y., 2010. The removal of copper (II) ion by using mushroom biomass (*Agaricus bisporus*) and kinetic modelling. Desalination 255 (1-3), 137–142.

Fadel, M., Hassanein, N.M., Elshafei, M.M., Mostafa, A.H., Ahmed, M.A., Khater, H.M., 2017. Biosorption of manganese from groundwater by biomass of *Saccharomyces cerevisiae*. HBRC J. 13 (1), 106–113.

Febrianto, J., Kosasih, A.N., Sunarso, J., Ju, Y.-H., Indraswati, N., Ismadji, S., 2009. Equilibrium and kinetic studies in adsorption of heavy metals using biosorbent: a summary of recent studies. J. Hazard Mater. 162 (2-3), 616–645.

Freundlich, H., 1906. Uber die adsorption in losungen, zeitschrift Fur phtsikalische chemie. Zeitschrift Fur Physikalische Chemie 62 (5), 121–125.

Ghaedi, M., Ghezelbash, G.R., Marahel, F., Ehsanipour, S., Najibi, A., Soylak, M., 2010. Equilibrium, thermodynamic, and kinetic studies on lead (II) biosorption from aqueous solution by Saccharomyces cerevisiae biomass. Clean 38 (9), 877–885.

Ghorbani, F., Younesi, H., Ghasempouri, S.M., Zinatizadeh, A.A., Amini, M., Daneshi, A., 2008. Application of response surface methodology for optimization of cadmium biosorption in an aqueous solution by *Saccharomyces cerevisiae*. Chem. Eng. J. 145 (2), 267–275.

Göksungur, Y., Üren, S., Güvenç, U., 2005. Biosorption of cadmium and lead ions by ethanol treated waste baker's yeast biomass. Bioresour. Technol. 96 (1), 103–109.

Hadiani, M.R., Darani, K.K., Rahimifard, N., Younesi, H., 2018a. Biosorption of low concentration levels of Lead (II) and Cadmium (II) from aqueous solution by *Saccharomyces cerevisiae*: response surface methodology. Biocatal. Agric. Biotechnol. 15, 25–34.

Hadiani, M.R., Khosravi-Darani, K., Rahimifard, N., 2019. Optimization of as (III) and as (V) removal by *Saccharomyces cerevisiae* biomass for biosorption of critical levels in the food and water resources. J. Environ. Chem. Eng. 7 (2), 102949.

Hadiani, M.R., Khosravi-Darani, K., Rahimifard, N., Younesi, H., 2018b. Assessment of mercury biosorption by *Saccharomyces cerevisiae*: response surface methodology for optimization of low Hg (II) concentrations. J. Environ. Chem. Eng. 6 (4), 4980–4987.

Khorasani, A.C., Shojaosadati, S.A., 2017. Improvement of probiotic survival in fruit juice and under gastrointestinal conditions using pectin-nanochitin-nanolignocellulose as a novel prebiotic gastrointestinal-resistant matrix. Appl. Food Biotechnol. 4 (3), 179–191.

Khosravi-Darani, K., Zoghi, A., Jazayeri, S., da Cruz, A.G., 2020. Decontamination OF aflatoxins with a FOCUS ON aflatoxin B1 BY probiotic bacteria and yeasts: a review. J. Microbiol. Biotechnol. Food Sci. 10 (3), 424–435.

Kumar, K.V., Sivanesan, S., 2006. Pseudo second order kinetic models for safranin onto rice husk: comparison of linear and non-linear regression analysis. Process Biochem. 41 (5), 1198–1202.

Langmuir, I., 1918. The adsorption of gases on plane surfaces of glass, mica and platinum. J. Am. Chem. Soc. 40 (9), 1361–1403.

Le, B., Yang, S.H., 2019. Biosorption of cadmium by potential probiotic *Pediococcus pentosaceus* using in vitro digestion model. Biotechnol. Appl. Biochem. 66 (4), 673–680.

Mahurpawar, M., 2015. Effects of heavy metals on human health. Int. J. Res Granthaalayah 2394–3629. ISSN-23500530.

Malik, L.A., Bashir, A., Qureashi, A., Pandith, A.H., 2019. Detection and removal of heavy metal ions: a review. Environ. Chem. Lett. 17 (4), 1495–1521.

Massoud, R., Hadiani, M.R., Hamzehlou, P., Khosravi-Darani, K., 2019. Bioremediation of heavy metals in food industry: application of *Saccharomyces cerevisiae*. Electron. J. Biotechnol. 37, 56–60.

Massoud, R., Khosravi-Darani, K., Sharifan, A., Asadi, G.H., Younesi, H., 2020. The biosorption capacity of *Saccharomyces Cerevisiae* for cadmium in milk. Dairy 1 (2), 169–176.

Massoud, R., Sharifan, A., Khosravi-Darani, K., Asadi, G., 2021. Mercury biosorption process by using *Saccharomyces cerevisiae* in milk. J. Food Process. Preserv. 45 (1), e15008.

Mello, L.C., Claudino, A., Rizzatti, I., Bortoluzzi, R.L., Zanette, D.R., 2005. Analysis of trace metals Cu2+, Pb2+ and Zn2+ in coastal marine water samples from Florianópolis, Santa Catarina State, Brazil. J. Braz. Chem. Soc. 16 (3a), 308–315.

Raikwar, M.K., Kumar, P., Singh, M., Singh, A., 2008. Toxic effect of heavy metals in livestock health. Vet. World 1 (1), 28.

Raoov, M., Mohamad, S., Abas, M.R., 2013. Removal of 2, 4-dichlorophenol using cyclodextrin-ionic liquid polymer as a macroporous material: characterization, adsorption isotherm, kinetic study, thermodynamics. J. Hazard Mater. 263, 501–516.

Ribeiro, V.R., Maciel, G.M., Fachi, M.M., Pontarolo, R., Fernandes, I.d.A.A., Stafussa, A.P., Haminiuk, C.W.I., 2021. Biosorption of biocompounds from white and green tea in *Saccharomyces cerevisiae* waste: study of the secondary metabolites by UPLC-QToF-MS and simulated in vitro gastrointestinal digestion. Food Biosci. 101001.

Savastru, E., Zamfir, C.-I., Diaconu, M., Bulgariu, D., Bulgariu, L., 2019. Biosorption of Cu (II) ions from aqueous solution on *Saccharomyces cerevisiae* biomass: isotherm and kinetics modelling. In: 2019 E-Health and Bioengineering Conference (EHB).

Savić, J.Z., Vasić, V.M., 2006. Thermodynamics and kinetics of 1, 8-dihydroxy-2-(imid-azole-5-ylazo)-naphthalene-3, 6-disulphonic acid immobilization on Dowex resin. Colloids Surf. A Physicochem. Eng. Asp. 278 (1-3), 197–203.

Sieuwerts, S., De Bok, F.A., Mols, E., De Vos, W.M., van Hylckama Vlieg, J., 2008. A simple and fast method for determining colony forming units. Lett. Appl. Microbiol. 47 (4), 275–278.

Soares, E.V., Soares, H.M., 2012. Bioremediation of industrial effluents containing heavy metals using brewing cells of *Saccharomyces cerevisiae* as a green technology: a review. Environ. Sci. Pollut. Control Ser. 19 (4), 1066–1083.

Tempkin, M., Pyzhev, V., 1940. Kinetics of ammonia synthesis on promoted iron catalyst. Acta Phys. Chim. USSR 12 (1), 327.

Tuzen, M., Sarı, A., Turkekul, I., 2020. Influential bio-removal of mercury using *Lactarius acerrimus* macrofungus as novel low-cost biosorbent from aqueous solution: isotherm modeling, kinetic and thermodynamic investigations. Mater. Chem. Phys. 249, 123168.

Wang, J., Chen, C., 2006. Biosorption of heavy metals by *Saccharomyces cerevisiae*: a review. Biotechnol. Adv. 24 (5), 427–451.

Water, S., Organization, W.H., 2006. Guidelines for Drinking-Water Quality [electronic Resource]: Incorporating First Addendum, 1. Recommendations.

Wu, Y., Wen, Y., Zhou, J., Dai, Q., Wu, Y., 2012. The characteristics of waste *Saccharomyces cerevisiae* biosorption of arsenic (III). Environ. Sci. Pollut. Control Ser. 19 (8), 3371–3379.

Yin, J., Wu, B., Liu, S., Hu, S., Gong, T., Cherr, G.N., Zhang, X.-X., Ren, H., Xian, Q., 2018. Rapid and complete dehalogenation of halonitromethanes in simulated gastrointestinal tract and its influence on toxicity. Chemosphere 211, 1147–1155.

Zinicovscaia, I., Grozdov, D., Yushin, N., Abdusamadzoda, D., Gundorina, S., Rodlovskaya, E., Kristavchuk, O., 2020. Metal removal from chromium containing synthetic effluents by *Saccharomyces cerevisiae*. Desalination Water Treat. 178, 254–270.

Zoghi, A., Khosravi-Darani, K., Sohrabvandi, S., 2014. Surface binding of toxins and heavy metals by probiotics. Mini Rev. Med. Chem. 14 (1), 84–98.

Zoghi, A., Khosravi _Darani, K., Hekmatdoost, A., 2020. Effects of pretreatments on patulin removal from apple juices using lactobacilli: binding stability in simulated gastrointestinal condition and modeling. Probiotics Antimicrob Proteins 1–11.