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Alizarin red-S removal from aqueous solutions using *Saccharomyces cerevisiae*: kinetic and equilibrium study

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ABSTRACT

Alizarin red-S (ARS) is a type of azo dye that is widely used for dyeing of wool and nylon. It is toxic and carcinogenic and can cause a serious hazard to aquatic ecosystems. The ARS removal was experimentally conducted using the *Saccharomyces cerevisiae* biomass under various parameters of pH (3–11), contact time (20–180 min), the ARS concentration (25–100 mg L^{-1}), and yeast dose (0.1%–1.3% w v^{-1}). After completing each run, the suspensions were centrifuged at 10,000 rpm for 7 min. Finally, the absorbance of samples was read by UV-visible spectrophotometer at the wavelength of 427 nm. The maximum removal efficiency of ARS (68.51%) occurred at the conditions of ARS concentration of 50 mg L^{-1}, *S. cerevisiae* dose of 0.4% w v^{-1}, solution pH of 3, and contact time of 120 min. Biosorption process is desired at low pH under acidic conditions. The equilibrium data of the biosorption process were well fitted with the Langmuir model (R^2 = 0.99). Results indicated that the removal efficiency of ARS has an increasing rate from 10 to 80 min. The best kinetic model to fit experimental data was the pseudo-second-order. The results of the current study confirmed that *S. cerevisiae* could be used as a low-cost biosorbent for eliminating ARS from aqueous solutions.

Keywords: Alizarin Red S; *S. cerevisiae*; Biosorption; Langmuir isotherm; Kinetic

1. Introduction

The dyes used in the textile industry are harmful and biologically stable compounds due to their aromatic rings. Furthermore, the discharge of industrial dyes to the water bodies even at low concentrations would lead to an increase in the amount of turbidity. The classification of dyes is based on their chemical nature as azo, anthraquinone, sulfur, triphenylmethane, and phthalocyanine compounds [1,2]. Among industrial dyes, alizarin red-S (ARS, $C_{14}H_7NaO_7S \cdot H_2O$, see Fig. 1) has been extensively used in the textile industry since ancient times. This type of dye is anionic and soluble in water, which is widely applied for dyeing of wool and nylon. Industrial dyes are often toxic and carcinogenic and can cause a serious hazard to aquatic organisms. Therefore, ARS has been selected as target pollutants in many investigations on the treatment of dye-laden wastewaters [3]. The treatment of dye wastewater was commonly carried out by physical and chemical processes like photocatalysis, ozonation, membrane filtration, electrocoagulation, and adsorption. These methods produce a large volume of sludge (which need to be managed) have a high cost and in some cases produce toxic by-products [1,4]. However, physicochemical processes (have extensive applications in dye decolorization, the biosorption and biodegradation by microbial biomass) are an eco-friendly and effective process to remove dye from wastewaters. In fact, biosorption and biodegradation approaches, compared with

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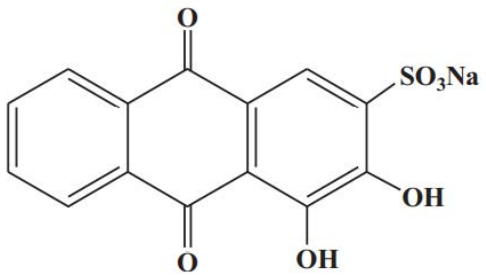

Fig. 1. Chemical structure of alizarin red S.

physical or chemical methods, can reduce energy requirements and the quantity and toxicity level of the resulted waste sludge which, in turn, leads to a further reduction of environmental impacts. In this regard, *S. cerevisiae* can metabolize and biosorb dye compounds [5–7]. This microorganism can be produced in large quantities and used as a by-product in some industries. In removal processes of dyes, microorganisms can adsorb and or degrade azo dyes into aromatic amines with side groups (–SO_3, –OH, –COOH, –Cl) [5,8]. Mc Mullan et al. reviewed the main mechanisms for dyestuffs removal by diverse categories of microorganisms from bacterial and fungal domains [8]. Vatandoostarani et al. and Jadhav et al. indicated that *S. cerevisiae* can completely remove the dye of methyl red at various times between several minutes to 24 h depending on the amount of cell mass used for the decolorization process [5,9]. Thus, in the current study, the removal of ARS by the *Saccharomyces cerevisiae* yeast was considered due to advantages such as safety, low cost, simplicity, widespread distribution, non-pathogenic, rapid growth of cells, and easy cultivation [10,11]. To the best of the authors' knowledge, there are no published papers about the ARS removal by using *S. cerevisiae*. The aim of this work was designed to examine the removal efficiency of ARS from aqueous solutions by *S. cerevisiae* and to evaluate biosorption capacity with different kinetic and isotherms models.

2. Materials and methods

2.1. Materials

The yeast of *S. cerevisiae* PTCC 5052 strain was provided from Iranian Research Organization for Science and Technology (IROST). In the experiment, the yeast was prepared at the concentration of 1%. For this purpose, 1 g of yeast was suspended in 100 mL of toxic substance solution. All chemicals applied in the experiments were of reagent grade. All solutions were prepared with distilled water. ARS was purchased from Merck (Darmstadt, Germany).

2.2. Preparing the reaction mixtures

The known amount of *S. cerevisiae* and 100 mL of the reaction mixture were poured in a series of 250 mL conical flasks and agitated at the fixed speed of 120 rpm in room temperature (28°C ± 2°C). The effects of the contact time (10–180 min), initial ARS concentration (25–100 mg L^{-1}), yeast dose (0.1%–1.3% w v^{-1}), and pH (3–11) were examined on the removal of ARS. To determine kinetic and isotherm models of the adsorption, 0.7 g of yeast was added to 100 mL of ARS solutions containing 25–100 mg L^{-1} of ARS, with the reaction times of 10–180 min, and the solution pH of 3.

After completing the removal, the resulting suspensions were centrifuged at 10,000 rpm for 7 min to remove the medium. Finally, the absorbance of each sample was read by UV-visible spectrophotometer, model T80+ (PG instrument Ltd., Leicester, UK), at the wavelength of 427 nm, and the amount of ARS in the samples was calculated based on the standard curve. To create the standard curve, all calibrations were made using four standards over the ARS range of 10–40 mg L^{-1} which resulted in the standard curve with the linear correlation coefficient (R^2) of 0.999. The removal efficiency (R, %) of ARS was calculated using the equations as follows:

$$R(\%) = \frac{(C_0 - C)}{C_0} \times 100 \quad (1)$$

where C_0 is the initial ARS concentration (mg L^{-1}) and C is the ARS concentration in solution after removal (mg L^{-1}). Fig. 2 indicates the removal of ARS using *S. cerevisiae* (0.7% w v^{-1}) with different concentrations of ARS.

2.3. Statistical analysis

All statistical analyses were performed with SPSS 16.0. Statistical analysis of the biosorption data was done via one-way ANOVA and Tukey HSD.

3. Results and discussion

3.1. Effect of independent variables on ARS removal

In this study, initially, the effects of the ARS concentration and *S. cerevisiae*, pH and the reaction time on the removal efficiency of ARS were investigated. Fig. 3 displays the combined effect of initial ARS concentration and contact time on ARS removal. The results presented in Fig. 3 show that there is a direct relationship between the removal efficiency of ARS and its initial concentration (P-values < 0.05). The highest removal rate was obtained at the initial ARS concentration of 75 mg L^{-1}, as the collision between *S. cerevisiae* and ARS increases. From the results, the curve slope initially increases and then gradually decreases along with the elevation of the

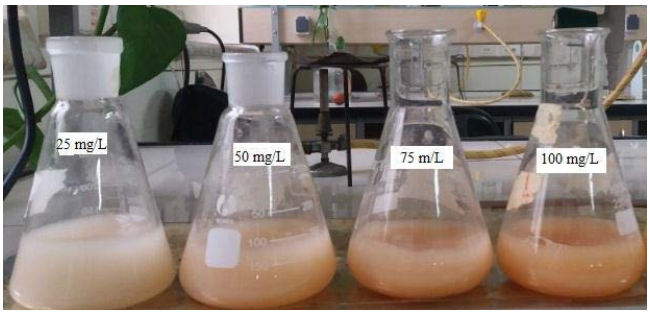

Fig. 2. Biosorption of ARS using wet cells of *S. cerevisiae* at an initial ARS concentration of 25–100 mg L^{-1}.

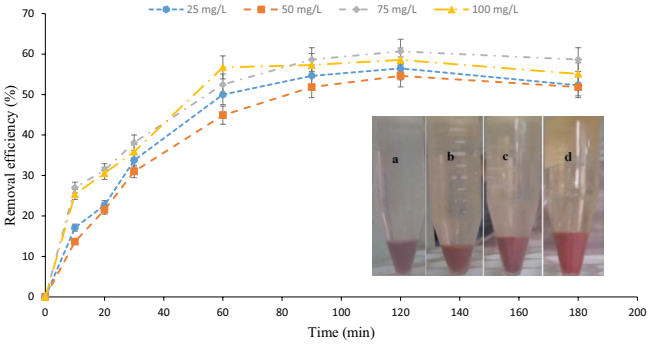

Fig. 3. The effect of the initial concentration of ARS on the removal efficiency at different contact time. Inset figure shows changes in ARS dye after centrifugation at the concentration of (a) 25 mg L^{-1}, (b) 50 mg L^{-1}, (c) 75 mg L^{-1}, and (d) 100 mg L^{-1}.

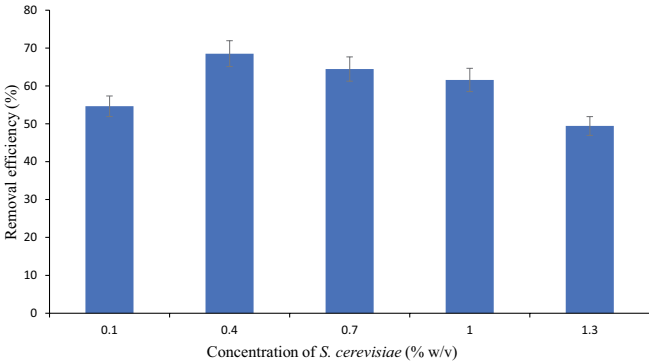

Fig. 4. The effect of *S. cerevisiae* concentration on the removal efficiency of ARS.

ARS concentration. The actual amount of the adsorbed dye increased with increasing the initial dye concentration from 25 to 75 mg L^{-1}. This was confirmed by other researchers [17,18] and they attributed this phenomenon to the increase in the driving force of the concentration gradient. Hence, a higher initial concentration of the dye will enhance the biosorption process. Then, the removal rate was gradually decreased with the increase in the ARS concentration from 75 to 100 mg L^{-1}. This is due to the biosorption capacity is influenced by the increase in the concentration of ARS which reduce the biosorption capacity owing to the saturating sorption sites of the cell wall of yeast [19]. Based on Fig. 3, there is a direct relationship between the removal efficiency of ARS and the reaction time (P-value < 0.05). The maximum removal efficiency of ARS was obtained 68.51% at the concentration of 0.4% *S. cerevisiae*. As seen from Fig. 3, ARS was removed rapidly from 0 to 60 min and after that from 60 to 180 min the curve slope was flatted. The change in the rate of biosorption might be due to the fact that initially, all the adsorbent sites are vacant and solute concentration gradient is very high. Later, the lower adsorption rate is due to a decrease in the number of vacant sites of adsorbent and dye concentrations. The decreased adsorption rate, particularly, toward the end of contact times indicates the possible monolayer formation of ARS on the adsorbent surface [20,21]. This may be attributed to the lack of available active sites required for further uptake after attaining the equilibrium [22].

Fig. 4 illustrates the effect of different concentrations of *S. cerevisiae* (0.1%–1.3% w v^{-1}) on the removal efficiency of ARS at the pH of 3 and the contact time of 60 min. Results (Fig. 4) show an increase in the uptake percentage of ARS with the increase in the concentration of *S. cerevisiae* from 0.1% to 0.4%. As can be observed from Fig. 4, by augmenting *S. cerevisiae* from 0.1% to 0.4%, the removal efficiency of ARS was increased from 54.6% to 68.5% and then decreases at *S. cerevisiae* concentrations of 0.4% to 1.3% (P-value > 0.05). The decrease in dye removal at high biosorbent dosages can be explained by referring to the aggregation of biosorbent particles to each other. Consequently, the accumulation and aggregation of biosorbent particles lead to a decrease in the total surface area of the biosorbent. Another reason may be due to the unsaturation of biosorption sites through the biosorption reaction [23].

Fig. 5 exhibits the effect of pH on the removal efficiency of ARS within 60 min of contact time. The results of this study (Fig. 5) reflect that with the increase in the solution pH from 3 to 11, the removal efficiency of ARS decreases (P-value < 0.05). The solution pH can change the surface charge and the ionization degree of materials [24]. The cell wall of yeast is influenced by the pH of the solution. Carboxyl, phosphonate, and amine groups play important roles in the biological activity of *S. cerevisiae*. Compared with other groups, amine groups are more active in absorbing contaminants due to creating positive charges on yeast. Carboxyl and phosphonate groups and phosphonate, on the other hand, bring negative charges on the yeast [24]. When the pH of the solution was approximately 3, *S. cerevisiae* would have been attraction of the dye molecule with the amino groups of *S. cerevisiae*. In aqueous solution, the ARS was first dissolved and the sulphonate groups of ARS ($D–SO_3Na$) dissociate and were converted to anionic dye ions ($D–SO_3Na \leftrightarrow D–SO_3^- + Na^+$). The amino groups of *S. cerevisiae* were then protonated under acidic conditions as follows: ($R–NH_2 + H^+ \leftrightarrow R–NH_3^{3+}$). In addition, under acidic conditions (pH < 3), the sulphonate groups ($D–SO_3^-$) combined with H^+, which decreased the adsorption capacity of ARS, according to the following reaction:($D–SO_3^- + H^+ \leftrightarrow D–SO_3H$). As a result, the sorption proceeded with electrostatic interactions between the two counterions ($R–NH_3^-$ and $D–SO_3^-$) as follows [15]: $R–NH_3^+ + D–SO_3^- \leftrightarrow R–NH_3^+…SO^3–D$. At pH 3, most of the $–NH_2$ groups were protonated, which were favorable for the adsorption of ARS [25]. However, at high pH, the number of protonated $–NH_2$ groups were decreased and more –OH

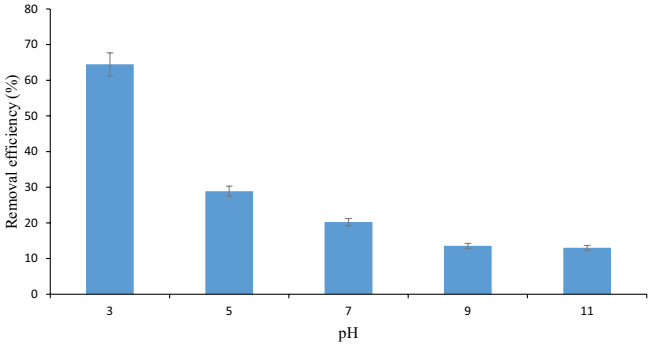

Fig. 5. The effect of initial pH on the removal efficiency of ARS.

were available to compete with the anionic sulphonic groups. As a result, the adsorption capacity for the ARS decreased at high pH [26]. The study of Kristina et al. (2008) confirmed that microorganisms could be having high removal efficiency at low pH [27]. Asku et al. (2003) used *S. cerevisiae* to remove reactive dye, where at a low pH, the removal efficiency of the reactive dye was significantly high [28].

3.2. Biosorption kinetics

The parameters of the kinetic model and regression correlation coefficient are presented in Table 1. In this study, the effect of adsorbent dose on the adsorption capacity (mg g^{-1}) was studied and the value of adsorption capacity was calculated by the kinetic reaction models, that is, the pseudo-first-order and pseudo-second-order models [29].

The pseudo-first-order model is expressed as Eq. 2:

$$\log\left(q_e - q_t\right) = \log q_e - \frac{k_1}{2.303} \times t \tag{2}$$

where k_1 (min^{-1}), q_e (mg g^{-1}), and q_t (mg g^{-1}) are the pseudo-first-order kinetic constant, adsorbed ARS per mass of *S. cerevisiae* at equilibrium, and the amount of adsorbed ARS per mass of the yeast at any time (min), respectively.

The pseudo-second-order model is described as follows:

$$\frac{t}{q_t} = \frac{1}{k_2 q_e^2} + \frac{1}{q_e} \times t \tag{3}$$

where k_2 (g mg^{-1}. min) is the rate constant of pseudo-second-order sorption. From Table 1, the pseudo-second-order model for studied concentrations of ARS was found more suitable for describing kinetic data according to the correlation coefficient ($R^2 \geq 0.97$) and obtained statistical data compared with pseudo-first-order model [30,31].

3.3. Biosorption isotherms

The Langmuir and Freundlich isotherm models were used to describe the biosorption equilibrium data.

The Langmuir isotherm equation is:

$$q_e = \frac{q_m K_L C_e}{1 + K_L C_e} \tag{4}$$

where q_e (mg g^{-1}), q_m (mg g^{-1}), K_L (L mg^{-1}), and C_e (mg L^{-1}) are expressed as the biosorption capacity at the equilibrium state, the maximum biosorption capacity, the Langmuir equilibrium constant, and the equilibrium concentration of ARS, respectively [32].

The linear form of the Freundlich model is presented as follows:

$$q_e = K_f C_e^{1/n} \tag{5}$$

where K_f and n reflect Freundlich constants which linked to the biosorption capacity and biosorption intensity, respectively [33].

Langmuir and Freundlich constants and correlation coefficients are listed in Table 2. As shown in Table 2, the Langmuir isotherm exhibited the best-fit model (R^2 = 0.99) for ARS removal, compared with the Freundlich model (R^2 = 0.97). Also, the maximum adsorption capacity of *S. cerevisiae* was obtained to be 29.41 mg g^{-1} based on the Langmuir model. Langmuir and Freundlich isotherms describe the mechanism of monolayer biosorption with homogenous energy and the multilayer biosorption with heterogeneous energy, respectively [34]. Table 3 demonstrates a comparison between the biosorption capacities of ARS dye with earlier studies. According to this table, the yeast biosorbent in this study has a good chance for competing with other adsorbents in the removal of dye.

Table 2
Langmuir and Freundlich parameters for the biosorption of ARS onto *S. cerevisiae*

Langmuir			Freundlich		
q_m (mg g^{-1})	K_L (L mg^{-1})	R^2	K_f (mg g^{-1})	1/*n*	R^2
29.41	0.006	0.99	0.14	1.09	0.97

Table 3
Comparison of maximum ARS biosorption capacity obtained in this work with previous studies

Sorbent	q_m (mg g^{-1})	Reference
S. cerevisiae	29.41	This work
Porous xerogels	8.30	[12]
Olive stone	16.01	[4]
MWCNT	161.03	[13]
Magnetic chitosan	40.12	[14]
Cynodon dactylon	16.32	[15]
Activated carbon	<20	[16]

Table 1
Parameters of kinetic model for ARS adsorption onto *S. cerevisiae* (pH: 3, yeast dose: 0.7%)

Alizarin red-S concentration (mg L^{-1})	Pseudo-first-order			Pseudo-second-order		
	R^2	q_e (mg g^{-1})	k_1 (min^{-1})	R^2	q_e (mg g^{-1})	k_2 (g mg^{-1} min)
25	0.88	0.93	0.72	0.97	2.27	0.018
50	0.83	0.93	1.07	0.97	4.76	0.001
75	0.82	0.93	1.27	0.99	7.14	0.008
100	0.83	0.92	1.41	0.98	9.09	0.007

4. Conclusion

In this paper, the removal of the ARS by the *Saccharomyces cerevisiae* yeast was studied. The results indicated that the maximum removal efficiency of ARS (68%) occurred at ARS concentration of 75 mg L^{-1}, biosorbent dose of 0.4 % w v^{-1}, pH 3, and contact time of 120 min. The ARS biosorption process was indicated to be dependent on pH, initial dye concentration, and contact time. The removal efficiency of ARS had an increasing rate from 10 to 180 min followed by equilibrium state. The dye biosorption process was desired under acidic conditions. The equilibrium biosorption data were better described by the Langmuir model. The pseudo-second-order model was the best model to fit the experimental data. The results of this study confirmed that *S. cerevisiae* could be used as a low-cost adsorbent for the removal of ARS from aqueous solutions.

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