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## Use of non-viable green alga *Chara* sp. for the removal of hazardous Alizarin dye from synthetic wastewater: Kinetics and Equilibrium Studies

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### Abstract

This work explores the use of non-viable green alga *Chara* sp. as a model potential biosorbent material, for anthraquinone dye alizarin sorption from synthetic wastewater. The impact of major variables like pH, biosorbent dose, contact time, initial dye concentration, etc. was investigated. The maximum adsorption was found at 180 min. contact time, 3pH, 4 g/L dose, and 318 K temperature. Maximum adsorption capacity (76.92mg/g) of alizarin dye on *Chara* sp. was observed at 200 g/L initial alizarin concentration. Various adsorption isotherm models (Langmuir, Freundlich, Tempkin and Dubinin-Radushkevich Isotherms) and kinetic models (pseudo-first-order, pseudo-second-order, and intraparticle models) were applied to analyze the adsorption data. Isotherms have also been used to determine thermodynamic parameters of the process, viz., free energy change, enthalpy change, and entropy change. The non-viable biomass was also investigated using scanning electron microscopy (SEM) and FTIR before and after alizarin dye adsorption. The results indicated that the biomass of *Chara* sp. is an effective and inexpensive biosorbent material for the removal of alizarin dye from synthetic wastewaters.

**Keywords:** Adsorption, *Chara* sp., Alizarin, Adsorption isotherms, kinetic models

### Introduction

It is difficult to treat dye-contaminated water due to its synthetic origin and complex aromatic molecules [1]. Several physical, chemical, and biological techniques have been developed over the years to eliminate hazardous dyes from aqueous solutions. Among various treatment

technologies, adsorption has been successfully employed for the decolorization of wastewater. The main advantages of an adsorption system for wastewater pollution control are less investment in terms of initial cost, immense effectiveness under favorable conditions, versatility, minimal volume of disposable sludge, and simple regeneration.

Several studies have been performed on the dye removal from water by various adsorbents viz. activated carbon, natural materials (clays, zeolites), and dead and living adsorbents (fungi, algae, and other microbial cultures) agricultural solid wastes (leaves, fibers, fruits peels, seeds, etc) [2-7] Decolorization of Alizarin dye using iron oxides was investigated by Pirillo et al. [8]. Similarly, Feng fu et al. have carried out a case study of adsorption of anionic dye Alizarin Red S on activated clay modified by iron oxide in a batch reactor [9]. Abdus et al. used an adsorbent prepared from mango seeds for the removal of Alizarin dye [10]. Similarly, the kinetics, adsorption isotherms, thermodynamics, and spectroscopic analysis of the removal of anthraquinone dye, Alizarin red S by adsorption onto mustard husk were studied by Gautam et al. [11].

In recent years, macroalga has attracted wide attention as a viable option to treat water-containing dyes [12]. Investigations conducted by several researchers demonstrated that *Chara sp.* is capable of accumulating dye like Lanaset Red G, Reactive Red (RR) 120 dye and Malachite green [13-15]. Still, there is lot of scope available to use this abundantly available alga for the removal of other dyes from wastewaters. Earlier, our lab utilized *Chara sp.* alga as an adsorbent for the removal of methylene blue dye from synthetic wastewater showing good adsorption capacity [16]. In the same sequence, in the present study, abundantly available green Charophyta alga, *Chara sp.*, was used as an adsorbent for the removal of Alizarin dye from synthetic wastewater. Synthetic wastewater used here, is the artificially prepared wastewater containing the desired concentration of the respective dye. The main aim of the present investigation is to identify the performance of non-viable alga *Chara sp.* on Alizarin dye removal from artificially prepared wastewater and to evaluate the effect of various parameters including contact time, pH, adsorbent dose, and temperature. Different adsorption

isotherms and kinetic models were also applied to fit the experimental data. The effectiveness of desorbing agents in stripping adsorbed dyes from biomass and its reuse was also investigated. This work, is our effort in continuation with previous studies on the removal of hazardous pollutants so as to make a green and pollutant-free atmosphere

## Materials and Methods

### Chemicals and Equipment

Technical grade Alizarin of 98% purity was used without further purification. All the other chemicals used in this study were of analytical grade. Measurements of pH were carried out on a digital pH meter (PERFIT, India) and absorbance of samples was recorded using UV-Vis Spectrophotometer-119 (Systronics India Ltd). The FTIR spectrum of biomass was determined using KBr pellets on a Thermo Nicolet FTIR (Germany) within 4000- 400  $\text{cm}^{-1}$  range. SEM was performed using ZEISS EVO 40 EP (Cambridge, UK) with analytical software Quantax 200 instrument. Elemental analyze system Vario MICRO CHNS V3.1.1 (GmbH, Germany) was used to carry out the analysis of Carbon, Sulphur and Nitrogen.

### Adsorbent and Adsorbate

The material upon whose surface the adsorption takes place is called adsorbent. For this study abundantly available green algae *Chara sp.* was utilized. They are multicellular and superficially resemble land plants because of stem and leaf-like structures (Fig.1). The collected biomaterial was then washed with tap water followed by distilled water several times to remove extraneous material. After Sun drying for two days, it was dried in the oven at 343 K for 24 hrs. The dried mass was ground using pestle mortar and sieved to the desired 100-micrometer mesh size and was maintained in vacuum desiccators prior to use.



Figure 1. Green algae *Chara* sp.

Alizarin dye was used as adsorbate in this study, is having a molecular formula  $C_{14}H_8O_4$ , chemical name 1,2-dihydroxyanthraquinone and molecular weight 240.21 g/mol . Figure 2 shows the molecular structure of Alizarin dye. The dye stock solution was prepared by dissolving accurately weighed amounts of dye in 1000 mL distilled water. Standard curve with excellent correlation

( $R^2 = 0.99$ ) was established for different concentrations of the dye solution (from 1 to 10 mg/L). The absorbance of dye solutions with known concentrations was measured by a Spectrophotometer at wavelength of 567 nm. Distilled water was used as a reference in all the experiments.

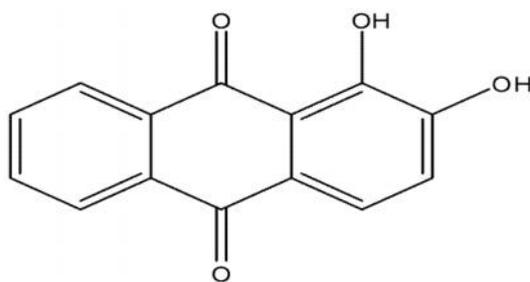


Figure 2. Structure of Alizarin dye

### Batch Studies

Adsorption experiments were performed in batch mode to investigate the effects of various parameters like contact time (0- 160 min), pH of solution (range 2- 12), adsorbent dose (1- 10 g/L) and temperature (298K, 308 and 318 K). To obtain adsorption isotherms, the adsorbent was

suspended in MO solution at three different temperatures i.e. 298, 308 and 318 K, while the other parameters were kept constant. Kinetic studies of adsorption were carried out at two different concentrations (100 and 200 mg/L) at 318 K wherein the extent of adsorption was analyzed at a regular time interval.

## Adsorption Isotherms and Kinetic Models

Table 1. Equations of various Adsorption Isotherms and Kinetic Models applied

Model	Equation	Parameters	References
<b>Langmuir Isotherm</b>	$\frac{1}{q_e} = \frac{1}{Q_0} + \frac{1}{bQ_0C_e}$	$q_e$ (mg/g): equilibrium adsorption capacity $Q_0$ (mg/g): maximum adsorption capacity $C_e$ (mg/L): equilibrium concentration of adsorbate $b$ : Langmuir constant	[17]
<b>Freundlich Isotherm</b>	$\ln q_e = \ln K_F + \frac{1}{n} \ln C_e$	$K_F$ ( $\text{mg}^{1-1/n} \text{L}^{1/n} \text{g}^{-1}$ ): Freundlich constant $n$ : intensity of adsorption.	[18]
<b>Temkin Isotherm</b>	$q_e = RTb_T \ln (A_T C_e)$	$R$ : gas constant ( $8.314 \text{Jmol}^{-1} \text{K}^{-1}$ ) $T$ : absolute temperature in Kelvin $b_T$ ( $\text{g kg mg}^{-1} \text{mol}^{-1}$ ) and $A_T$ ( $\text{L mg}^{-1}$ ): constant associated with the heat of adsorption	[19]
<b>D-R Isotherm</b>	$\ln q_e = \ln q_m - \frac{2}{RT} \ln (1+1C_e)$	$q_m$ (mg/g): maximum adsorption capacity $E$ ( $\text{mol}^2 \text{kJ}^{-2}$ ): constant related to adsorption energy $\omega$ : Polanyi potential	[20]
<b>Kinetic Study</b>			
<b>Pseudo-first order</b>	$\log (q_e - q_t) = \log q_e - \frac{k_{1,ads}}{2.303} t$	$q_t$ (mg/g): amount of dye adsorbed at equilibrium time $t$ $k_1$ ( $\text{min}^{-1}$ ): pseudo-first-order rate constant.	[21]
<b>Pseudo-second order</b>	$\frac{t}{q} = \frac{1}{k_{2,ads} q_e^2} + \frac{1}{q_e} t$	$q_e$ (mg/g): amount of the dye adsorbed at equilibrium $q$ (mg/g): amount of the dye adsorbed at time $t$ $k_2$ ( $\text{g mg}^{-1} \text{min}^{-1}$ ): rate constant of second order adsorption	[22]
<b>Intra particle model</b>	$q_t = k_{id} t^{1/2}$	$q_t$ (mg/g): amount of dye adsorbed per unit mass of adsorbent at time $t$ $k_{id}$ ( $\text{mg/g min}^{-1/2}$ ): intra-particle diffusion rate constant	[23]

## Results and Discussion

Characterization of *Chara sp.*

The Surface area of the algal biomass *Chara sp.* was found to be  $0.855 \text{m}^2/\text{g}$ , by BET method.

Elemental Analysis of algal biomass showed the composition of Carbon, Hydrogen Nitrogen and Sulphur to be 13.55, 0.477, 0.32 and 0.426% respectively, Carbon being the prominent component of algal biomass.

## SEM Micrographs

SEM images (Figure 3a, & b), show a clear difference in the morphology of algal biomass before and after adsorption of Alizarin dye. The images revealed that alga has a massy thick

covering showing a number of amorphous particles scattered on the surface (figure 3b). Thus, it is clear that the dye molecules have been trapped and adsorbed on the active sites of the adsorbents.

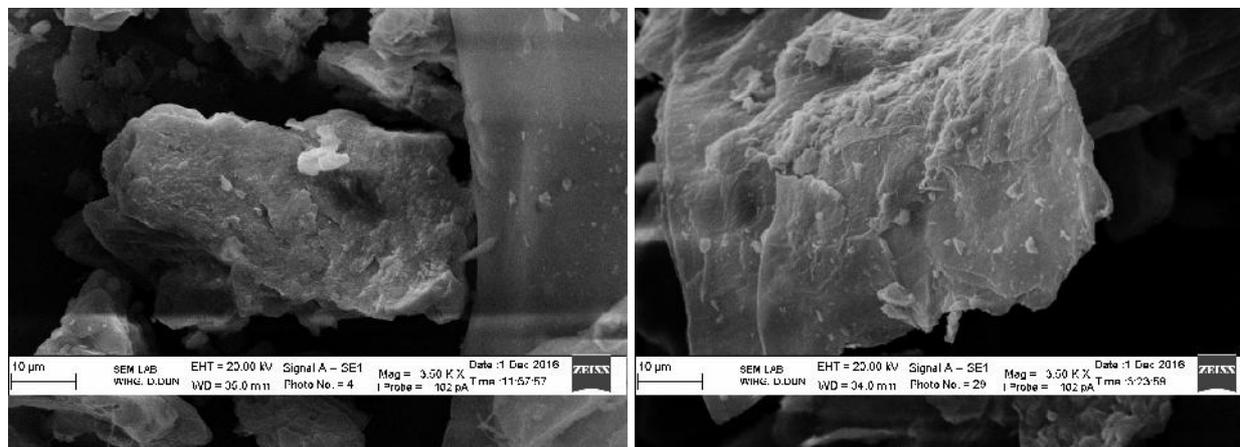


Figure 3. SEM micrographs of (a) *Chara sp.* before adsorption (b) *Chara sp.* after adsorption of Alizarin dye

## FTIR studies

FTIR investigation was performed to obtain information on the possible interactions between the functional groups of algal biomass surface and dyes. The FTIR spectra of algal biomass before and after dye removal are shown in Figure 4a and 4b. Comparison of dye loaded biomass with

spectra of unloaded biomass displays significant changes in the peaks (Table 2), indicating the presence of several functional groups on the surface of algal biomass that were responsible for binding with dye. Similar results were reported for the removal of Lanaset Red G on macro alga *Chara contraria* [13].

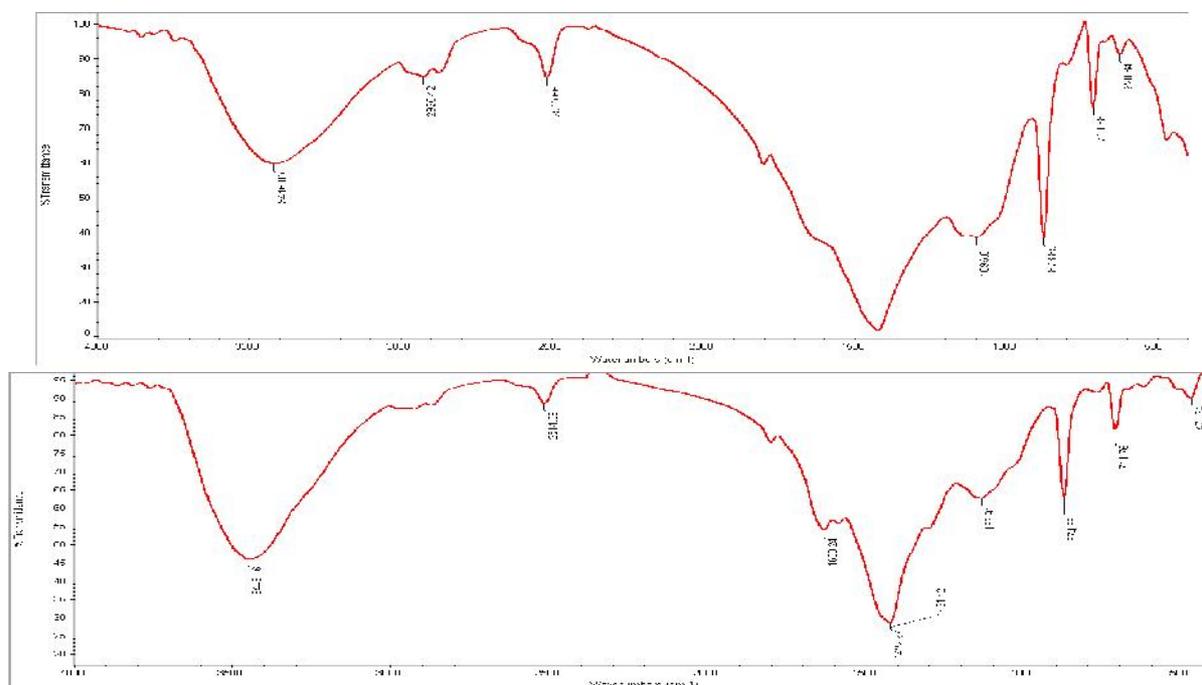


Figure 4 FTIR spectra of *Chara sp.* (a) before adsorption (b) after adsorption of Alizarin dye

**Table 2 IR adsorption band and corresponding possible functional groups present on *Chara sp.* surface.**

Before adsorption of Alizarin dye (cm <sup>-1</sup> )	After adsorption of Alizarin dye (cm <sup>-1</sup> )	Bonds indicative of functional groups
3416.05	3448.16	Carboxylic/OH stretch and N-H stretch
2515.66	2514.69	Phenolic/ carboxylic
1096.81	1133.91	C-N<
873.85	874.76	C-N-S scissoring

### Optimized parameters studied

Adsorption experiments were performed in batch mode to investigate the effects of few parameters such as contact time, pH, adsorbent dosage and temperature. To study the equilibration time for the maximum adsorption of the Alizarin dye on *Chara sp.*, the adsorption at four different concentrations (50, 100, 150, and 200 mg/L) were studied.

#### Effect of contact time

The adsorption of Alizarin dye onto *Chara sp.* increases as the contact time increases. The perusal of Figure 5 (a) reveals that the curve is smooth and continuous and reaches equilibrium at 180 min, suggesting possible monolayer coverage of Alizarin on *Chara sp.* After a rapid diffusion of dye molecules from bulk solution to exterior surface of adsorbent particles, the removal rate slowed down gradually until the equilibrium reaches. Similar results were reported by Karthik et al. for the removal of synthetic dye effluents by sago waste [24].

#### Effect of pH

Figure 5(b) depicts the effect of pH on the adsorption of Alizarin on the algal biomass. The dye uptake decreased with an increase in pH. The observed decrease in the uptake at low pH 3 is attributed to the decrease in Alizarin dissociation, which led to a lower concentration of anionic dye species available to interact with active sites of

the adsorbent. Above the optimum pH value of 3.0, the algal biomass displayed a sharp decrease in the uptake as at higher pH the extent of protonation gets lowered [25].

#### Effect of adsorbent dosage

The perusal of Figure 5 (c) depicts the amount of adsorbent significantly influencing the adsorption of Alizarin by *Chara sp.* At a dose of 4g/L, *Chara sp.* showed maximum adsorption. This may be attributed to the fact that at a low dose all the type of active sites is exposed and adsorption occur faster and attains saturation. But after a certain limit, when the dose is increased, the partial overlapping or aggregation of adsorbent arises, thus, resulting in decrease in the total surface area and availability of adsorption sites [26].

#### Effect of temperature

As the temperature increased from 298 to 318 K, an increase in the adsorption process occurs, as shown in figure 5(d), which indicates its endothermic nature. As the temperature increases, the boundary layer thickness of the adsorbent decreases thus leading to the increase in the availability of active sites and porosity and also kinetic energy of dye molecules [27]. The endothermic nature of dye sorption has also been reported for the adsorption of Reactive Red 120 dye onto *Chara contraria* [14].

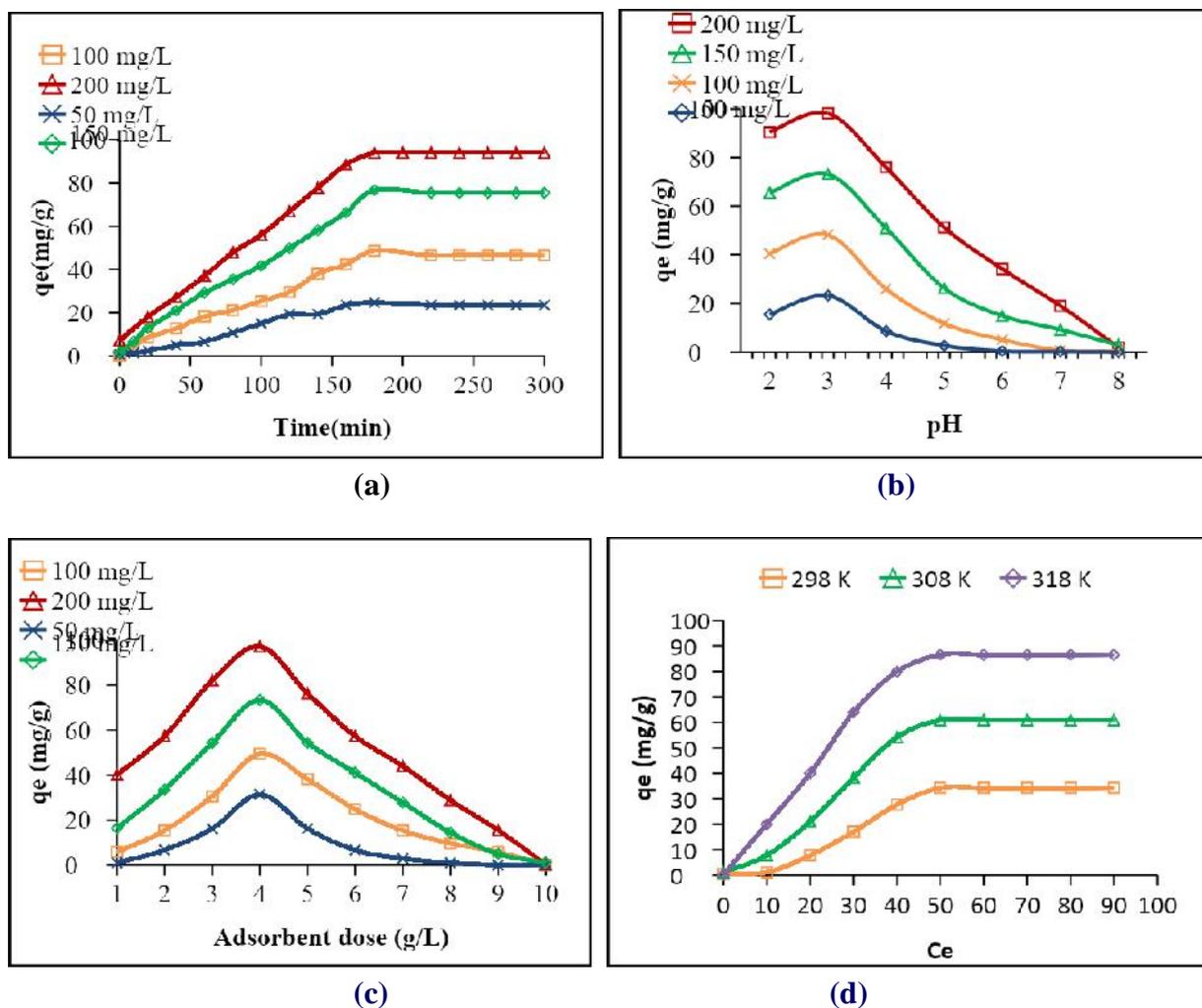


Figure.5 Effect of (a) contact time (b) pH (c) adsorbent dose (d) temperature on the adsorption of Alizarin dye onto *Chara* sp.

### Isotherm modeling of adsorption

To analyze the validity of the adsorption data, Langmuir, Freundlich, Temkin and Dubinin-Radushkevich adsorption models were run at three different temperatures (298, 308 and 318K). The calculated values of adsorption isotherm

parameters and correlation coefficient ( $R^2$ ) for Alizarin adsorption onto *Chara* sp. are listed in Table 3. As it can be seen the values of correlation coefficient were high at different temperatures for Langmuir followed by Freundlich model and then Temkin and D-R models respectively.

Table 3 Isotherm parameters for the adsorption of Alizarin dye onto *Chara sp.*

Isotherm Parameters	Alizarin		
	298 K	308 K	318K
<b>Langmuir Isotherm</b>			
<b>b (L mg<sup>-1</sup>)</b>	0.036	0.041	0.044
<b>q<sub>e</sub> (mg g<sup>-1</sup>)</b>	55.55	70.07	76.92
<b>R<sup>2</sup></b>	0.991	0.990	0.999
<b>Dimensionless Separation Factor</b>			
<b>R<sub>L</sub></b>	0.008	0.076	0.066
<b>Freundlich Isotherm</b>			
<b>N</b>	1.102	1.253	1.432
<b>K<sub>F</sub> (mg g<sup>-1</sup>)</b>	2.295	3.068	4.997
<b>R<sup>2</sup></b>	0.929	0.935	0.963
<b>Temkin Isotherm</b>			
<b>A<sub>T</sub></b>	0.803	0.869	0.948
<b>b<sub>T</sub></b>	64.01	74.91	100.45
<b>R<sup>2</sup></b>	0.909	0.946	0.927
<b>D-R isotherm</b>			
<b>q<sub>m</sub> (mg g<sup>-1</sup>)</b>	63.50	79.33	91.14
<b>E (kJ mol<sup>-1</sup>)</b>	1.17	1.29	1.58
<b>R<sup>2</sup></b>	0.917	0.910	0.927

### Thermodynamic Parameters

Table 4. represents the values of changes in Gibbs free energy(  $G^{\circ}$ ), enthalpy (  $H^{\circ}$ ), and entropy (  $S^{\circ}$ )for the adsorption of Alizarin dye onto *Chara sp.* These were calculated using the following equations.

$$\Delta G^{\circ} = RT \ln(b) \quad (2)$$

$$\ln\left(\frac{b_2}{b_1}\right) = -\frac{\Delta H^{\circ}}{R} \left(\frac{1}{T_2} - \frac{1}{T_1}\right) \quad (3)$$

$$\Delta G^{\circ} = \Delta H^{\circ} - T\Delta S^{\circ} \quad (4)$$

The decrease in  $G^{\circ}$  values with increasing temperature indicates the increase in feasibility and spontaneity of the adsorption at a higher temperature. The positive values of  $H^{\circ}$  and  $S^{\circ}$  indicates the endothermic nature of adsorption and the increased randomness at the solid/solution interface respectively. Daneshwar *et al.* have reported similar results for the removal of Acid Blue 25, Acid Orange 7 and Acid Black 1 dyes by brown macroalga *Stoechospermum marginatum* [28].

Table 4. Thermodynamic Parameters for the adsorption of Alizarin dye onto *Chara sp.*

Dye	Thermodynamic parameters	298 K	308 K	318 K
Alizarin	<b>G<sup>o</sup></b>	-16.757	-17.704	-24.517
	<b>S<sup>o</sup></b>	0.0894	0.08957	0.1081
	<b>H<sup>o*</sup></b>	9.886		

\*  $H^{\circ}$  Measured between 298 and 318K

### Kinetic studies

From Table 5, it can be inferred that pseudo-second-order adsorption model is more suitable for describing the adsorption kinetics of dyes onto

*Chara sp* in terms of higher values of correlation coefficient ( $R^2$ ) at two different concentrations. Similar results have been earlier reported by Nemr et al. for the removal of Chrysophenine dye by *Ulva lactuca* green alga [29].

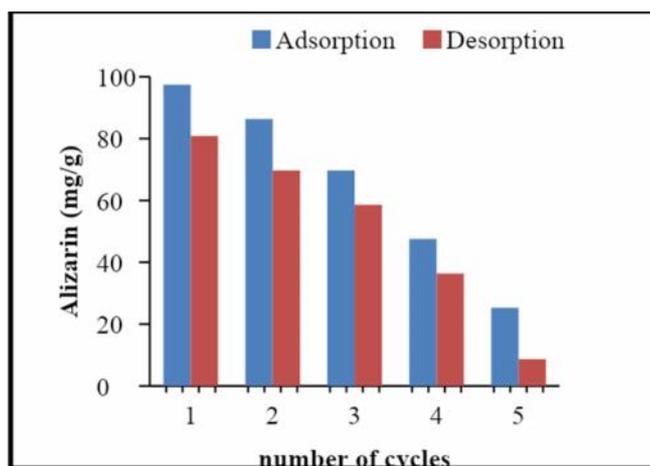
**Table 5. Kinetic parameters estimated for the adsorption of Alizarin dye onto *Chara sp*.**

Dye	Initial dye conc. (mg/L)	First-order model			Second-order model			Intra-particle model	
		$K_1(\times 10^{-3} \text{ min}^{-1})$	$q_e$ (mg $\text{g}^{-1}$ )	$R^2$	$K_2 \times 10^{-3}$ (g $\text{mg}^{-1} \text{ min}^{-1}$ )	$q_e$ (mg $\text{g}^{-1}$ )	$R^2$	$K_w$ (mg $\text{g}^{-1} \text{ min}^{0.5}$ )	$R^2$
Alizarin	100	9.212	85.901	0.979	0.201	90.90	0.990	3.117	0.936
	200	34.54	86.099	0.975	0.162	96.15	0.991	4.859	0.936

### Adsorption Desorption and Reuse studies

The effect of various reagents used for desorption studies shows that Hydrochloric acid is a better reagent for desorption. Stirk and Staden also found that acids are more effective for desorbing as compared to other eluents [30]. The desorption efficiency decreases with an increase in the number of cycles due to the decrease in the

adsorption capacity as shown in figure 6. The adsorption capacity of the tested alga did not noticeably change (only a maximum 20-25% change was observed) during repeated adsorption-desorption operations. Thus, the reuse of the biomass is an important feature for its possible utilization in the continuous system in industrial processes.



**Figure 6. Adsorption/Desorption cycles of Alizarin dye on *Chara sp* using HCl**

### Conclusions

The maximum adsorption capacity of Alizarin onto *Chara sp* was found to be 76.92 mg/g at pH 3, contact time 180 min, dose 4g/L, and temperature 318 K. The Langmuir isotherm was

found to be the best fit with the equilibrium experimental data followed by Freundlich, Temkin, and D-R isotherms. Thermodynamic parameters showed that the adsorption of Alizarin dye onto algal biomass was feasible, spontaneous, and endothermic under studied conditions.

The sorption kinetics followed a pseudo-second-order rate equation. The interaction/binding between the dye molecules and the functional groups on the cell wall surface of the biomass was confirmed by FTIR analysis, which indicated the participation of carboxylic, amino, amide, and hydroxyl functional groups in the dye adsorption. The adsorbent could be reused for five consecutive adsorption and desorption cycles with a negligible decrease in their adsorption capacities. Thus, *Chara* sp. algal biomass seems to be a promising biosorbent material for the treatment of alizarin dye – enriched wastewater.

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### **Conflict of interest:**

The authors declare that there is no conflict of interest.

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