Indian Journal of Chemical Technology
Vol. 29, September, 2022 pp. 511-518
DOI: 10.56042/ijct.v29i5.53698

Adsorptive column studies for removal of Reactive Blue 4 dye using dry cells of *Rhizopus oryzae* (MTCC 262)

Mukulika Bagchi, Debabrata Bera & Sunita Adhikari (Nee Pramanik)*

Department of Food Technology and Biochemical Engineering, Jadavpur University, Kolkata 700 032, India

E-mail: sunitapramanik@gmail.com

Received 12 August 2021; accepted 4 July 2022

The biosorption potential of dry biomass of *Rhizopus oryzae* (MTCC 262) in removing a textile dye Reactive Blue 4 from dye contaminated wastewater using up-flow packed-bed column reactor has been studied. The impact of operating parameters like flow rate of dye solution, bed height of column and concentration of dye in the feed solution on the removal potential has also been studied. The Adam-Bohart Model, Yoon-Nelson Model and Thomas model are used for the analysis of experimental data. Dye removal of 71.81% is obtained using dry biomass at flow rate of 2 mL $min^{-1}$of dye solution, bed height of 15 cm and influent dye concentration of 200 mg L^{-1}. Adsorbed dye can be successfully recovered from dry *R. oryzae* (MTCC 262) biomass of packed bed column reactor by running 1 (N) NaOH solution. Results demonstrate significant reduction in Reactive Blue 4 concentration after the biosorption process.

Keywords: Biosorption, Isotherm models, Packed bed column, Reactive Blue 4, *Rhizopus oryzae* (MTCC 262)

Textile industries are one of the most significant users of dyes among all industrial sectors. The textile industrial processing is very water-consuming which involves extensive use of synthetic dyes for dyeing process[1]. Apart from textile industries, dyes have wide applications in other industries viz., tannery, food, paper and pulp, printing, carpet and mineral processing etc.[2]. Dyes are common contaminants in industrial wastewater, which are potentially carcinogenic, mutagenic and allergenic to aquatic plants and animals. The existence of dyes in wastewater is visible easily due to their characteristic colour even when present in small concentrations[3,4]. The most common dyes are non-biodegradable easily because of their synthetic origin and complex aromatic structure. It can stay in the environment for a prolonged time period[5]. So, exclusion of dyes from the wastewater of textile industry is a foremost environmental issue of concern.

For large scale industrial effluent treatment packed bed column along with continuous flow operations are more efficient because it makes better use of the driving force that is concentration difference for adsorption. It allows more efficient utilization of the adsorbent capacity[6,7]. In a packed bed column industrial effluent is passed through it, that result in a better quality of effluent which can be discharged into aquatic streams. Using a defined quantity of sorbent in a packed bed column, large volume of wastewater can be treated continuously[8]. Various adsorbents like bagasse fly ash for adsorption of Acid orange dye[9], radiation-induced grafted fibers (AFF) for the removal of acid blue 80 (AB 80)[10], chitosan–glutaraldehyde biosorbent for Direct Blue 71 dye[11], *Rhizopus arrhizus* as a biosorbent for removal of remazol[12], *Bacillus* sp. for removal of Brilliant Red HE-3B textile dye[13] was studied by various investigator in column adsorption process. However, little effort has been employedby afew researchers for the studies on adsorption of dyes in a packed bed column [14-18]. Also it is successfully used in the process of cyclic sorption and desorption for maximum utilization of bio sorbent.

In present study, the treatment of Reactive Blue 4 solution using dry *Rhizopus oryzae* (MTCC 262) in a packed bed column reactor was studied. The reactor is an up-flow type. During the experiments bed height of the bed was varied, flow rate of the dye solution and dye concentration was also varied. The ratio of final and initial effluent dye concentration was measured and it was plotted against time. It was usually referred as breakthrough curve and used to describe the performance of the packed bed column[7,19,20]. Different models like Adam-Bohart (AB) model, Yoon and Nelson (YN) model and modified Thomas model were tested to fit the experimental data. The

breakthrough curve for the process of adsorption was predicted from it.

Desorption of the loaded dye is important at regular interval from the dye laden packed bed of the column for making the column operation successful. Desorption ensures the utilization of the same packed bed for a number of times; therefore, it minimizes the operational cost, excessive sludge generation and enhancing column efficiency. As performed in batch process, here also desorption was carried out using a base sodium hydroxide. Desorption of Reactive Blue 4 from dry *R. oryzae* biomass was carried out using 1 (N) NaOH solution as desorbing agent.

The process of packed bed adsorption has several advantages with respect to the process engineering. The operation is simple producing more yield. The process of scaling up from a laboratory scale set up is easy. Automation of the different stages of the separation process can be done. In a single step process often the high degree of purification can be achieved. For the proper design of continuous flow sorption processes studies on packed columns are necessary. It allows utilization of the sorbent more efficiently and provides better quality of the effluent for the dye biosorption process. Use of continuous packed bed column reactor with dry biomass was reported by various authors. Many mathematical models were used for the study of packed bed system and dynamic behaviour. The objective of this study is to study the removal of the dye Reactive Blue 4 from aqueous solution in a packed bed up-flow column reactor using dry cells of *Rhizopus oryzae* (MTCC 262) biomass. In this study the effects of various design parameters like flow rate, bed height, and inlet concentration of the dye solution were investigated. In addition, the rate-limiting step for the process and the kinetics of biosorption process were also studied.

Experimental Section

Fungal strain and maintenance

The fungal strain *Rhizopus oryzae* (MTCC 262) that was used in this study was purchased from the Institute of Microbial Technology, Chandigarh, India. Potato dextrose slant was used as the maintenance medium for the organism by sub culturing at 30°C for 120 h maintaining a regular interval of 30 days.

Medium composition and production of biomass

Medium composition for maintenance of biomass was as follows (potato dextrose agar): (g L^{-1}); potato extracts: 200.0, dextrose: 20.0, agar: 20.0, *p*H 5.0. The same medium without agar was used as the inoculum and growth medium. For preparation of inoculum one loopful of biomass from slant culture was transferred to 50 mL inoculum medium taken in a 250 mL Erlenmeyer flask. The flasks were incubated at 30°C, 120 rpm for 48 h. Then 1 mL of the inoculum was added to 50 mL of growth medium taken in a 250 mL Erlenmeyer flask and incubated for 72 h at 30°C for biomass production.

Preparation of dry cells

After completion of fermentation process the broth was centrifuged at 5000 rpm for 15 min to harvest the fungal mycelia. It was then washed thrice with distilled water and dried by lyophilization. Afterwards using a mixer grinder, the dried biomass was ground and used for adsorption study.

Chemicals

The dye Reactive Blue 4 used in this study was purchased from Sigma-Aldrich Chemical Co., St. Louis, MO, USA. Other chemicals and the ingredients of microbiological media were procured from E. Merck, Germany and Hi Media, India.

Preparation of dye solution and its estimation

A stock solution of the RB4 dye having concentration of 1000 ppm was made by mixingthe dye in distilled water. It was diluted again to obtainrequired concentration of dye for the experiments. The dye solution concentration was measured using UV–vis spectrophotometer (HITACHI U-2000) at 595 nm (λ_{max}).

Experimental design

The process of dye exclusion was examined on a lab scale up-flow type packed bed column reactor. Figure 1 schematically shows experimental setup of

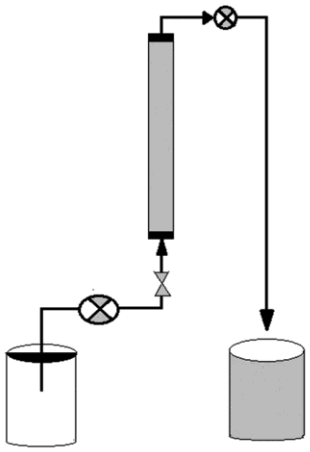

Fig. 1 — Schematic diagram of packed bed column reactor

the reactor. In a glass column (35 cm in length internal diameter 0.95 cm) the experiments were carried out. Packed bed of variable height was prepared with dry *R. oryzae* (MTCC 262) biomass. The effect of different parameters like influent dye concentration, inlet flow rate and bed height were studied. The experiments were carried out at actual bed height of 10 cm, 15 cm and 20 cm. The inlet flow rate of the dye solution was changed in the range of 2 mL min^{-1} to 4 mL min^{-1}. The experiments were carried out at inlet dye concentration of 50 mg L^{-1}, 100 mg L^{-1} and 200 mg L^{-1}. The concentration of dye in the effluent at fixed interval of time was continuously monitored for adsorption of dye by the packed bed and prediction of the breakthrough curve.

The breakthrough curve for adsorption of a solute from its solution into packed bed column under fixed operational conditions was expressed in terms of adsorbed dye concentration that is ($C_{ad}=C_0-C$) or it is expressed as a ration of final and initial dye concentration (C/C_0) that is a function of time or effluent volume, V_{eff}[21]. Where the preliminary dye concentration was C_0 and the ultimate dye concentration was C at time (t).

The effluent volume was calculated from equation (1).

$$V_{eff} = Q_{total} \quad \ldots(1)$$

In the above equation Q was the inlet flow rate of the dye solution (mL min^{-1}) and the total flow time (min) was expressed by t_{total}.

If the adsorbed dye concentration (C_{ad}) was plotted against time and integrating C_{ad} (mg/L) against t (min) the area under the curve (A) in mg/ min/L. At a given dye concentration, the quantity of total adsorbed dye (q total in mg) and flow rate was calculated from equation (2).

$$q_{total} = QA/1000 \int C_{ad}\, dt \quad \ldots(2)$$

Using equation (3) the total quantity of dye that passed through the column (m_{total}) in mg and it was calculated as

$$m_{total} = (C_0 \times Q_{total}) / 1000 \quad \ldots(3)$$

Total percentage (% removal) of dye removal was calculated from the following equation (4).

$$\% \text{ removal} = (q_{total}/ m_{total}) \times 100 \quad \ldots(4)$$

Where, W_h can be calculated from equation (5)

$$W_h = (\pi\, d^2_{column}) \times H/4 \quad \ldots(5)$$

The internal diameter of the column is d_{column} and effective bed height is H

Desorption experiments

The adsorption experiment was carried out using previously mentioned glass column at optimized flow rate (2 mL min^{-1}) and at the bed height of 20 cm for dry *R. oryzae* packed bed respectively and at initial dye concentration of 100 mg L^{-1}.

The desorption experiment was performed by passing 1 (N) NaOH solution through the column packed with dry *Rhizopus oryzae* cells in the up-flow mode at a speed of 2 mL min^{-1}. After each adsorption desorption the bed of the column was washed with distilled water. The adsorption-desorption was repeated for four consecutive cycles.

The regeneration studies indicated that the dry biomass of *Rhizopus oryzae* could be used repeatedly for four repeated cycles of adsorption-desorption to treat the Reactive Blue 4 containing effluent as some amounts of biomass were lost during the phase alteration (adsorption-desorption) of each cycle. After fourth cycle, the adsorption percentages reach below 50 percent.

Surface characteristics of the adsorbent

For FTIR spectra analysis, 0.1 of KBr was mixed with approximately 0.01 g of dry fungal biomass. By pressing with the aid of a bench press this mixture was pressed into a tablet form. FTIR Spectra of pristine and dye loaded *R. oryzae* (MTCC 262) biomass were obtained using IR spectrophotometer (Shimadzu Corporation, IR-Prestige 21, resolution 4 cm^{-1}). The spectrum was analyzed at a frequency of 200 kHz (i.e. 0.0805 s/scan, 40 s total for an experiment) in the region of 4000-400 cm^{-1}.

Using a JEOL, JMS 5600 (Japan) scanning electron microscope SEM of the pristine and dye loaded dry biomass of *R. oryzae* were obtained after coating with thin layer platinum under reducedpressure.

Results and Discussion

Effect of flow rate

The flow rate of feed solution (Q) was changed from 2 to 4 mL min^{-1} the effect of flow rate was studied at a bed height of 15 cm with dry biomass and 50 mg L^{-1} of dye concentration (*p*H 3.0). The flow of dyesolution through the dry biomass in the column was allowed for 360 min. The bed became saturatedwith dye within this time period. The solute concentration in the effluent will reach to C_0 i.e., the

system had reached the breakpoint. With increasing flow rate the adsorption of dye using dry biomass was found to decrease and shown at Fig. 2(a). With increasing flow rate at a fixed bed height, the breakpoint time decreased. At higher flow rate much sharper breakthrough curves were obtained as because the dye residence time in the column was not extended enough to attain the adsorption process equilibrium[22,23]. The reduction in percentage removal for higher flow rate could be due to insufficient contact time between adsorbent and dye[24]. With increase in flow rate of dye solution the dye removal was found to decrease into the column and maximum dye removal of 67.26% was observed at 2 mL min^{-1} flow rate with the dry biomass of *R oryzae* (MTCC 262) as per Table 1.

Effect of bed height of column

The column bed height (H) was varied from 5 to 15 cm for the dry biomass of *R oryzae* (MTCC 262) at a flow rate of 2 mL min^{-1} while the other experimental conditions were kept unchanged. With the dry biomass the maximum dye biosorption was observed at a bed height of 15 cm. The effect of bed height on biosorption of dye was shown in Fig. 2(b). With growing bed height at a fixed flow rate the breakpoint time rises. With the increase in bed height the total number of binding sites increased as a result in the increase of the amount of biomass that enhances the process of adsorption[6,22] due to rapid mass transfer[25]. As bed height increases, surface area of adsorbent also increased that provided more binding sites for adsorption. Using the dry biomass maximum dye removal of 72.96% was obtained at a bed height of 15 cm as depicted in Table 2.

Effect of initial dye concentration of the feed solution

For this study the initial dye concentration of the feed solution (C_0) was varied from 50 to 200 mg L^{-1} with the bed height of 15 cm for dry biomass and the other conditions were kept unchanged. The packed bed was saturated at lower influent dye concentration after comparatively exposed for a long period of time. At lower dye concentration the curve of dye adsorbed (mg g^{-1}) versus time (t) was found to be dispersed in nature as shown in Fig. 2(c). The curve was much sharper at higher inlet dye concentration. Effect of initial dye concentration was related to the available sites present on the surface of adsorbent. With increase in initial dye concentration the breakpoint time decreased. This is because thebetter concentration gradient between the dye solution and solid matrix that offers a driving force for the speedy binding site saturation in the column[22]. With the increase in initial dye concentration adsorption capacity increased due to high driving force of mass transfer with initial high concentration of dye[26]. At 200 mg L^{-1} of influent dye concentration maximum dye removal of 71.81% was observed shown in Table 3 for dry biomass.

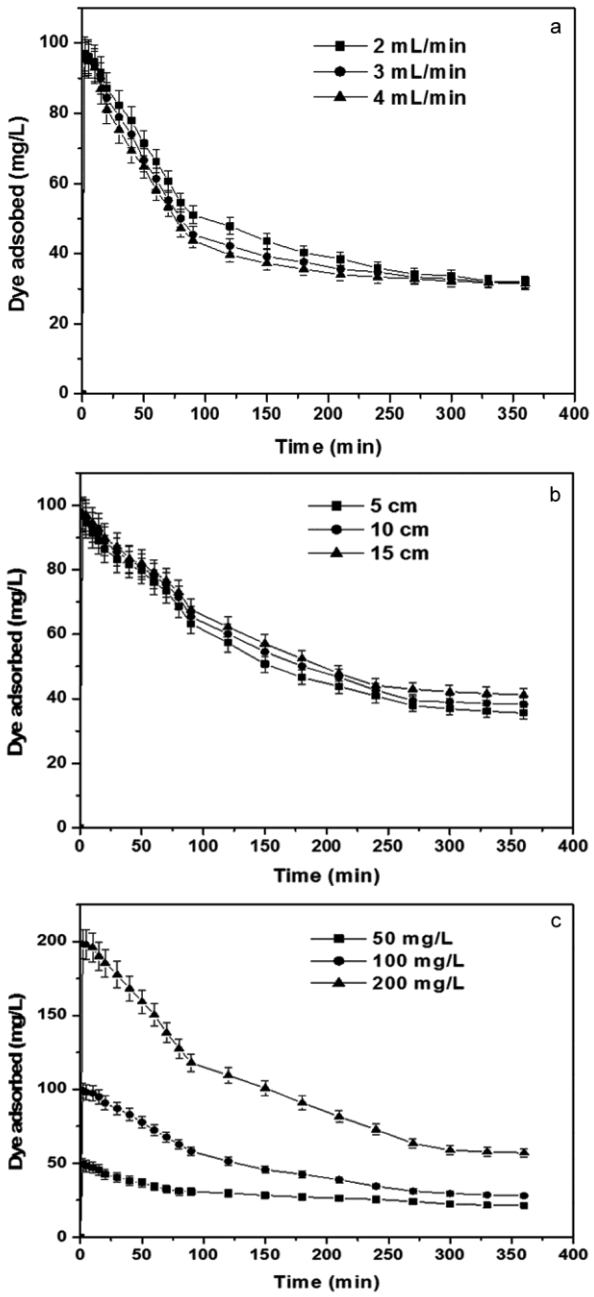

Fig. 2 — Effect of (a) flow rate; (b) bed height and (c) initial dye concentration on adsorption of Reactive Blue 4 by dry biomass of *R. oryzae* using packed bed column

Table 1 — Effect of flow rate on percentage removal of Reactive Blue 4 using dry biomass (Bed height: 15 cm, dye concentration: 100 mg L^{-1}, *p*H: 3.0).

Flow rate (mL min^{-1})	A (mg $min^{-1}L^{-1}$)	q_{total} (mg)	m_{total} (mg)	% Removal
2	13910.04	26.78	41	67.26
3	10774.26	33.46	62	57.88
4	9894.32	39.88	85	50.42

Table 2 — Effect of Bed height on percentage removal of Reactive Blue 4 using dry biomass of *R. oryzae* (MTCC 262) (flow rate: 2 mL min^{-1}, dye concentration: 100 mg L^{-1}, *p*H: 3.0)

Bed height (cm)	A ($mg^{-1}min^{-1}L^{-1}$)	q_{total} (mg)	m_{total} (mg)	%Removal
5	12268.26	24.66		59.92
10	12067.19	27.04	40	66.7
15	14108.42	28.92		72.96

Table 3 — Effect of dye concentration on percentage removal of Reactive Blue 4 with dry biomass of *R. oryzae* (Flow rate: 2 mL min^{-1}, bed height: 15 cm, *p*H: 3.0)

Dye concentration (mg L^{-1})	A (mg min^{-1} L^{-1})	q_{total} (mg)	m_{total} (mg)	% Removal
50	7671.57	23.69	39	61.7
100	10198.17	45.74	85	70.42
200	118994.29	64.45	97	71.81

Application of the Adam-Bohart model

Adams and Bohart first established a relationship between C/C_0 and t for a gas charcoal adsorption system[27]. However for any other adsorption processes this concept can be used. The initial part of the breakthrough curve is described by the Adam Bohart model. From the bulk solution through the bead material diffusion of solute occurs to the core of bead. Initially the concentration of solute in core area is negligible, so rate of diffusion follow a direct connection with time. Slowly the solute concentration increases in the core area. So the rate of diffusion declines and finally it becomes constant. For this model the mass transfer rates follow the subsequent equation (6).

$$\ln C/C_0 = K_{AB}C_0t - K_{AB}N_0(H/Q) \quad \ldots(6)$$

Where, C_0 and C are the inlet and outlet (mg L^{-1}) dye concentration respectively. K_{AB} is the kinetic constant (L $mg^{-1}min^{-1}$), N_0 is the saturation concentration (mg L^{-1}). The bed height is H (cm) and flow rate is Q (mL min^{-1}) .

Parameters that describe the characteristic operation of the column (K_{AB} and N_0) were determined and it was used for linear regression analysis. The K_{AB} and N_0 values were determined from the slope and intercept of the linear plot of ln C/C_0 against time (t). A linear relationship between C/C_0 and t was obtained after applying Equation (6) to the experimental data at different experimental conditions. From which the respective values of K_{AB} and N_0 were calculated for dry biomass of *R. oryzae* (MTCC 262). With rise in initial dye concentration as well as the flow rate the value of kinetic constant (K_{AB}) decreased but with rise in flow rate and dye concentration N_0 increased. This indicated that at the initial stage of adsorption in the column the overall system kinetics was dominated by external mass transfer[21]. A simple approach for evaluating the adsorption process was provided by Adam-Bohart model, though its validity is restricted to the range of conditions used[27-29]. For initial part of the breakthrough curve the correlation coefficient (R^2) values were found between 0.972 and 0.982 for dry biomass and presented in Table 4.

Application of Yoon and Nelson model

A comparatively simple model regarding gas-charcoal adsorption was developed by Yoon and Nelson[30]. This model is less complicated than other models. It does not need detailed data about the adsorbate characteristics, adsorbent type and the physical characteristics of the bed. In the form of Equation (7) this model is expressed.

$$\ln [C/(C_0-C)] = K_{YN}t - \tau K_{YN} \quad \ldots(7)$$

Where, the rate constant (min^{-1}) is expressed as K_{YN}. The time required for 50 % adsorbate break through time is expressed as τ.

The values of K_{YN} and τ were determined from slope and intercept of the respective linear regression fit from a linear plot of ln $[C/(C_0-C)]$ against sampling time (t) for dry biomass and presented in Table 5. The

Table 4 — Adam-Bohart parameters at different conditions using linear regression analysis

Flow rate (mL min^{-1})	Dye concentration (mg L^{-1})	K_{AB} (L mg^{-1} min^{-1})	N_0	R^2
	50	0.00012	2145.68	0.978
2	100	0.000048	3569.17	0.980
	200	0.000078	5684.14	0.982
3	100	0.00006	3767.34	0.978
4	100	0.000034	5805.94	0.972

Table 5 — Yoon and Nelson parameters at different conditions using linear regression analysis

Flow rate (mL min^{-1})	Dye concentration (mg L^{-1})	K_{YN} (min^{-1})	τ (min)	R^2
	50	0.0044	472.4	0.906
2	100	0.0065	466.2	0.943
	200	0.0088	510.6	0.946
3	100	0.0042	398.5	0.967
4	100	0.008	361.2	0.955

Table 6 — Thomas model parameters at different conditions using linear regression analysis

Flow rate (mL min^{-1})	Dye concentration (mg L^{-1})	K_{TH} (mL $mg^{-1}min^{-1}$)	q_0 (mg g^{-1})	R^2
	50	0.097	20.44	0.929
2	100	0.062	42.65	0.955
	200	0.045	66.87	0.962
3	100	0.547	10.14	0.973
4	100	0.425	11.24	0.966

correlation coefficient (R^2) values were found between 0.906 and 0.967. With the increase in flow rate it was observed that the value of K_{YN} decreased but it increased again with further rise in inlet concentration of the dye.

Thomas model

The plug flow nature in the bed has been adopted in this model. It uses second-order reversible reaction kinetics and Langmuir isotherm. For the adsorption process this model is appropriate in the absence of limitations of external and internal diffusion processes. For the successful design of the process of column adsorption the extreme adsorption capacity of an adsorbent is necessary. Traditionally to fulfill the purpose the Thomas model is used. In Equation (8) the linear form of Thomas model[31] is stated.

$$\ln [(C_o/C)-1] = (K_{Th}q_0M - k_{Th}C_0V_{eff})/Q \quad \ldots(8)$$

Where, K_{TH} is the Thomas rate constant (mL $min^{-1}mg^{-1}$), q_0 (mg g^{-1}) is the uptake capacity of the adsorbent at equilibrium, $C_0$is the inlet and C is the outlet dye concentration (mg L^{-1}), M (g) is the quantity of adsorbent and Q (mL min^{-1}) is the flow rate.

To obtain the respective linear regression fit the experimental data were fitted to the modified Thomas model. Using linear regression analysis the determined coefficients and relative constants were obtained as per Equation (8) and the results are listed in Table 6**.** As the influent dye concentration increased the value of K_{TH} decreased. These could be attributed to the increase in driving force with the increase in influent dye concentration[16].

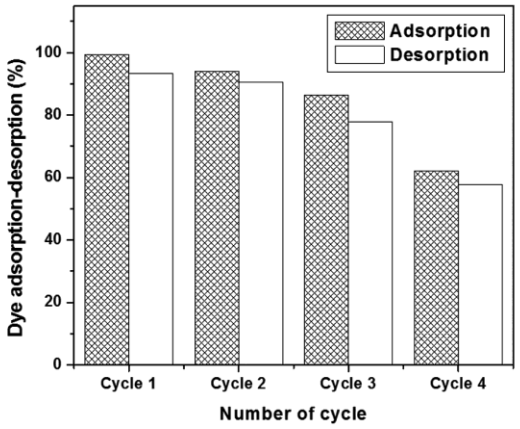

Fig. 3 — Adsorption-desorption cycle of Reactive Blue 4 using dry biomass of *Rhizopus oryzae* (MTCC 262)

Desorption of Reactive Blue 4 from column reactor packed with dry biomass of *R. oryzae*

In this case it is seen that after the 4th adsorption-desorption cycle, 57.78% of dye can be desorbed (Fig. 3) using dry biomass of *R. oryzae* (MTCC 262) in packed bed column reactor.

Surface characteristics of the adsorbent

FTIR was performed to get a knowledge related to the probable adsorption mechanism by detecting the

functional groups present on the cell surface as each group has unique energy absorption band. FTIR spectrum of pristine *Rhizopus oryzae* biomass exhibited distinct peaks suggesting the presence of amine, carbonyl, phosphate and hydroxyl groups as shown in Fig. 4. The sharp peak at 1646 cm^{-1} can be credited to the stretching of carboxyl or amide groups of C=O group. The band at 1550 cm^{-1} was due to bending of N–H bond. The peak position at 1452 cm^{-1} and 1400 cm^{-1} on the biomass could be attributed to COO− of the carboxylate. The peak at 3404 cm^{-1} due to amino group has been shifted to 3409 cm^{-1} after dye adsorption. Peak shape has been altered and peak

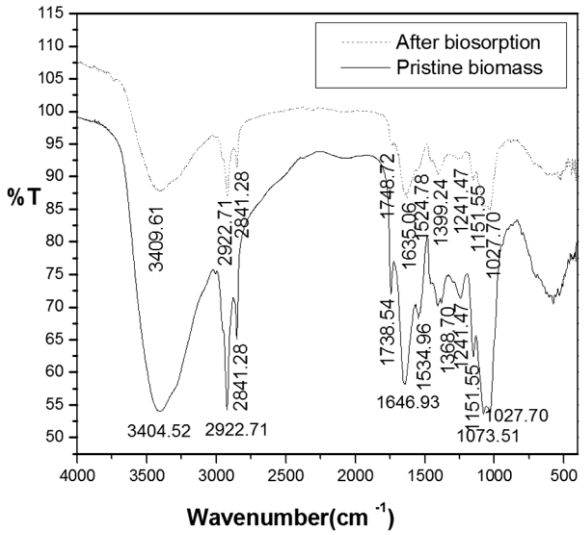

Fig. 4 — FTIR spectroscopy study of pristine biomass and dye loaded biomass of *Rhizopus oryzae* (MTCC 262)

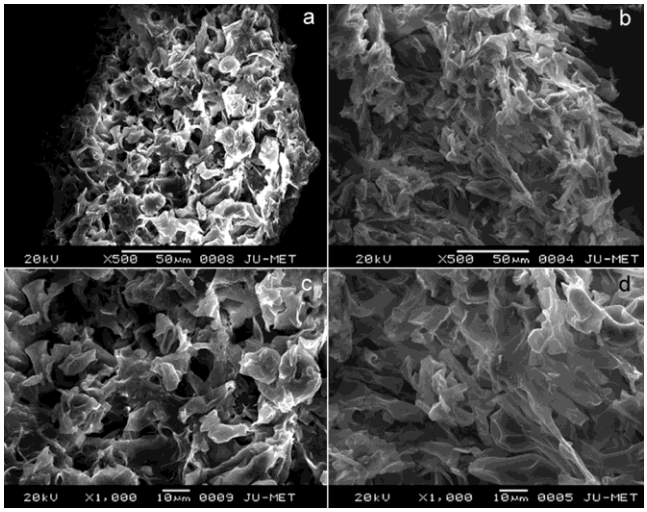

Fig. 5 — Scanning electron micrographs of *Rhizopus oryzae* (MTCC 262) biomass: (A) pristine biomass, (B) dye loaded biomass, (C) pristine biomass at higher magnification and (D) dye loaded biomass at higher magnification

at 1738 cm^{-1} had been shifted to 1748 cm^{-1} due to carboxylate group in pristine biomass. Due to due to involvement of C=O of carboxyl or amide group peak of 1646 cm^{-1} in pristine biomass had been also shifted to 1635 cm^{-1}. These results indicated the role of amino and carboxylate group on the process of dye adsorption.

Using scanning electron microscopy as a tool the surface morphology of a biosorbent can be extensively characterized. The surface morphology of the pristine *Rhizopus oryzae* biomass are expressed in Fig. 5 (A), (C). It appeared as irregular and rough structure with large surface area for interaction of dye molecule. After dye adsorption as per Fig. 5 (B), (D) significant changes were observed in surface morphology. It depicted that the dye had been adsorbed throughout the surface and along the cell boundary edge.

Conclusion

For optimum removal of dye through packed bed column using dry biomass it is needed to optimize some process parameters like bed height, flow rate and influent dye concentration. The data obtained by varying bed height, flow rate and dye concentration in a series of experiments were fitted to different models in order to predict the breakthrough curve of the adsorption. The fitted models were Adam-Bohart model, Yoon and Nelson model and the modified Thomas model. With lower dye concentration, lower feed flow rate and higher bed height the packed bed adsorption system was found to perform better. Removal of Reactive Blue 4 of 71.81% was obtained using dry biomass at the flow rate of 2 mL min^{-1}, bed height of 15 cm and influent dye concentration of 200 mg L^{-1}. At the initial part of breakthrough curve Adam–Bohart model fitted well whereas the other two models showed poor fitting to the breakthrough curve.

To perform a successful dye removal procedure, it is not only necessary to develop a continuous packed-bed column bioreactor system with a powerful adsorbent material for onsite operations, but also to determine the dye recovery percentage with an efficient desorbent solution[3]. Adsorbed dye can be successfully recovered from dry *R. oryzae* (MTCC 262) packed bed column reactor by running 1 (N) NaOH through the bed and it showed up to 57.78% of dye recovery after fourth adsorption-desorption cycles. The eluants desorb dye without significant deterioration of adsorption capacity of the bed. As recovery of the adsorbed dye molecules is possible to

a considerable extent from the bed so it can be concluded that dry *R. oryzae* packed bed column may be repeatedly utilized for practical purposes for the removal of Reactive Blue 4.

Acknowledgement

The authors would like to acknowledge DST, Government of India for providing the monetary support to carry out this researchand Department of Food Technology and Biochemical Engineering, Jadavpur University, India to conduct the experimental studies.

Disclosure statement

There is no conflict of interest.

References

1 Spagni A, Grilli S, Casu S & Mattioli D, *Int Biodeter Biodegr*, 64 (2010) 676.
2 Bagchi M & Ray L, *Chem Spec Bioavailab*, 27 (2015) 112.
3 Nath J, Das A & Ray L, *Ind Chem Eng*, 57 (2015) 82.
4 Nigam P, Armour G, Banat I M, Singh D & Marchant R, *Bioresour Technol*, 72 (2000) 219.
5 Safa Y, Bhatti H N, Bhatti I A & Asgher M, *Can J Chem Eng*, 89 (2011) 1554.
6 Adhikari S, Chattopadhyay P & Ray L, *Chem Spec Bioavailab*, 24 (2012) 167.
7 Adhikari S, Chattopadhyay P & Ray L, *Chem Spec Bioavailab*, 25 (2013) 273.
8 Aksu Z, Egretli G & Kutsal T, *Pros Biochem*, 33 (1998) 393.
9 Deokar A, Patel H, Thakare P, Bhagat S, Gedam V V & Pathak P, *Indian J Chem Technol*, 28 (2021) 319.
10 Selambakkannu S, Nor Azillah F O, Khomsaton A B & Zulhairun A K, *S N Appl Sci*, 1 (2019) 175.
11 Lopez-Cervantes J, Sanchez-Machado D & Sanchez-Duarte R, *Adsorp Sci Technol*, 36 (2018) 215.
12 Saraf S & Vaidya V K, *Am Inst Chem Eng Environ Prog*, 37 (2018) 355.
13 Horciu L, Zaharia C, Blaga A C, Rusu L & Suteu D, *Appl Sci*, 11 (2021) 4498.
14 Chen K C, Wu J Y, Huang G C, Liang Y M & Hwang S C J, *J Biotechnol*, 101 (2003) 241.
15 Fu Y & Viraraghavan T, *Water*, 29 (2003) 465.
16 Padmesh T V N, Vijayaraghavan K, Sekaran G & Velan M, *Dyes Pigments*, 71 (2006) 77.
17 Saeed A, Iqbal M & Zafar S I, *J Hazard Mat*, 168 (2009) 406.
18 Vijayaraghavan K & Yun Y S, *Chem Eng J*, 145 (2008) 44.
19 Cheng J & Wang X, *Sep Purif Technol*, 19 (2000) 157.
20 Ekpete, O A, Horsfall M J, Tarawou T, *J Appl Sci Environ Manage*, 15 (2011) 141.
21 Aksu Z & Gonen F, *Pros Biochem*, 39 (2004) 599.
22 Maity S K, Bera D & Ray L, *Appl Biochem Biotechnol*, 159 (2009) 488.
23 Robinson T, Chandran B, Naidu G S & Nigam P, *Bioresour Technol*, 85 (2002) 43.
24 Deokar S, Patel H, Thakare P, Bhagat S, Gedam V V & Pathak P, *Indian J ChemTechnol*, 28 (2021) 319.
25 Selambakkannu S, Othman N A F, Bakar K A & Karim Z A, *S N Appl Sci*, 175 (2019) 2910
26 Soltani A, Faramarzi M & Parsa S A M, *Water Quality Res J*, 56 (2021) 181.
27 Bohart G & Adams E Q, *J Am Chem Soc*, 42 (1920) 523.
28 Chu K H & Hashim M A, *J Environ Sci*, 19 (2007) 928.
29 Chafi M, Akazdam S, Asrir C, Sebbahi L, Barka N G & Essahli M, *Inter J Material Text Eng*, 9 (2015) 1242.
30 Yoon Y H & Nelson J H, *AIHA Journal*, 45 (1984) 509.
31 Thomas H G, *Ann NYA*, 49 (1948) 161.