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Polysulfone-immobilized *Corynebacterium glutamicum*: A biosorbent for Reactive black 5 from aqueous solution in an up-flow packed column

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ABSTRACT

Polysulfone-immobilized *Corynebacterium glutamicum* was employed as a biosorbent, for the continuous removal of Reactive black 5 (RB5) from aqueous solution, in an up-flow packed column. The biosorbent performance was evaluated with different bed heights (8–10 cm), flow rates (0.5–1 ml/min) and initial dye concentrations (50–100 mg/l). Favorable conditions for RB5 biosorption were observed with the highest bed height (10 cm), lowest flow rate (0.5 ml/min) and lowest initial dye concentration (50 mg/l); at which the RB5 uptake and % removal, 88.9 mg/g and 61.8%, respectively, were recorded. Mathematical modeling of experimental data was performed, using a non-linear form of the Thomas, modified dose–response and Yoon–Nelson models, to simulate the breakthrough curves. Very favorable results were obtained with the Thomas and Yoon–Nelson models, which described the experimental data well, with very high correlation coefficients. In an attempt to regenerate the exhausted biosorbent for possible reuse in multiple cycles, 0.1 M NaOH was employed as elutant. Due to continuous usage of polysulfone-immobilized *C. glutamicum* in three sorption–desorption cycles, a decreased breakthrough time, increased exhaustion time, broadened mass transfer zone, flattened breakthrough curve and decreased RB5 uptake were observed with progressive cycles. Linear regression of the breakthrough, uptake and critical bed length revealed that the sorption zone would reach top of the bed after 18 cycles, with the column bed completely exhausted after 35 cycles. The elutant, 0.1 M NaOH, provided uniform elution efficiencies greater than 99.2% in all three cycles.

1. Introduction

Environmentally, the presence of synthetic dyes in wastewaters is of particular concern due to their toxicity to some aquatic organisms and serious health risk to humans [1], leading to greater public concern and present legislation problems. Hence, there is a need to develop an effective process that can efficiently treat dye-bearing wastewaters. The search for new and innovative treatment technologies has focused attention on the dye-binding capacities of biological materials. Inactive/dead biomass of various bacteria [2], fungi [3], fresh water algae [4,5] and seaweeds [6], as well as other biosorbents [7,8] have been proposed as materials capable of sorbing different dyes from aqueous solutions. The mechanisms associated with dye biosorption by biological materials are often complex and involve both extracellular and intracellular dye binding. Amongst the biosorbents used in dye removal, microorganisms, such as bacteria and fungi, have been portrayed to possess excellent dye-binding capacities [9,10]. However, these types of biosorbents are usually soft and poses problem in solid–liquid separation after biosorption [11]. Desirably, the biosorbent must be hard enough to withstand the application pressures, as well as porous and regeneratable. Researchers have recognized that immobilizing biomass in a granular or polymeric matrix may improve the performance of the biomass and facilitate its separation from solutions [10,11]. The important matrices used in biosorbent immobilization include polysulfone [12,13], polyacrylamide [14], polyurethane [15] and sodium alginate [14]. Polysulfone is an amorphous, rigid, heat-resistant and chemically stable thermoplastic material, which has been identified as a good immobilizing agent [16]. The choice of immobilization matrix is a key factor in the environmental application of immobilized biomass. The polymeric matrix determines the mechanical strength and chemical resistance of the final biosorbent particles for successive biosorption cycles [14].

Continuous-flow columns are often used in industry for the treatment of dye-bearing wastewaters. Packed [5,16] and fluidized columns [17] have also been used for continuous dye biosorption. In process applications, a packed bed column is an effective arrangement for cyclic sorption–desorption, as it makes the best use of the concentration difference, which is known to be a driving force for

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biosorption, resulting in a better quality of effluent [18,19]. However, only little effort has focused on employing packed columns for dye biosorption; Fu and Viraraghavan [16] employed a glass column (1.27 cm i.d. and 40 cm height) packed with polysulfone-immobilized *Aspergillus niger* biomass for the removal of Acid blue 29, Basic blue 9, Congo red and Disperse red 1 from aqueous solutions. Padmesh et al. [5] used a glass column (2 cm i.d. and 35 cm height) packed with *Azolla filiculoides* for the treatment of Acid blue 15 contaminated solutions.

For a successful biosorption technology, the biosorbent must have a high dye-binding capacity and also be abundantly available so as to ensure a continuous supply for the process [20]. Fermentation industries usually generate huge amounts of waste biomass of microbial origin. *Corynebacterium glutamicum* biomass, which exhibited excellent dye biosorption capacity in our previous studies [2,21], is generated in huge quantities in amino acid fermentation industries.

Therefore, this study aimed to investigate the removal of Reactive black 5 (RB5), a model dye solute, from aqueous solution, using polysulfone-immobilized *C. glutamicum* in an up-flow packed column. Experiments were conducted as a function of the bed height (8–10 cm), flow rate (0.5–1 ml/min) and inlet dye concentration (50–100 mg/l). In addition, regeneration experiments were conducted to explore the possible reuse of the biosorbent.

2. Experimental

2.1. Solute and preparation of biosorbent

RB5 ($C_{26}H_{21}N_5Na_4O_{19}S_6$), with a purity and molecular weight of 55% and 991.82, respectively, was purchased from Sigma–Aldrich, Korea Ltd. (Yongin, Korea).

The fermentation wastes (*C. glutamicum* biomass) were obtained in a dried powder form from a lysine fermentation industry (BASF-Korea, Kunsan, Korea), and were grounded and sieved to obtain particle sizes in the range of 0.1–0.25 mm. The biomass (10 g/l) was then protonated with 0.1 M HNO_3 for 1 h at room temperature (25 °C). The biomass, after pretreatment, was washed with deionized water until the pH of the wash solution was approximately 7.0. The wet biomass was then dried in an oven at 60 °C for 12 h. A 9% (w/v) solution of polysulfone was prepared in *N,N*-dimethyl formamide (DMF) solution. After stirring the mixture for 10 h, the protonated biomass (14%) was mixed with the polysulfone slurry, with the resultant slurry dripped in deionized water, where beads were formed by a phase inversion process. The beads were washed with deionized water, and then placed in a water bath for 18 h to remove all residual DMF. The resultant beads (1–2 mm diameter) were stored at 4 °C.

2.2. Column arrangement and procedure

A glass column (1 cm i.d. and 12 cm height) was packed with a known quantity of the biosorbent to yield the desired bed height. The column was then fed with a known concentration of RB5 solution (pH 1), in an up-flow mode, at the desired flow rate using a peristaltic pump. Samples were collected at the exit of the column at desired time intervals, using a sample collector arrangement, and the dye concentration analyzed using a spectrophotometer (UV-2450, Shimadzu, Kyoto, Japan) at 597 nm. The column operation was stopped when the effluent dye concentration exceeded 95% of that at the inlet.

In the regeneration experiments, the dye-loaded biosorbent was exposed to 0.1 M NaOH, at a flow rate of 1 ml/min. After elution, deionized water was pumped through the column to wash the bed until the pH in the wash water stabilized near to 7.0. The regenerated bed was reused in the next cycle. These cycles of biosorption followed by elution were repeated three times to evaluate the biosorbent capacity. To determine the weight loss after three cycles, the biosorbent was washed with deionized water and dried naturally.

3. Theory

3.1. Analysis of column data

To represent the dynamic dye removal of packed columns, a mathematical analysis of the system was performed, and S-shaped experimental curves (C/C_0 vs. time) evaluated. The breakthrough time (t_b) was represented as the time at which the outlet RB5 concentration reached 1 mg/l, and the exhaustion time (t_e) the time at which the outlet RB5 concentration exceeded 95% of that at the inlet.

The total quantity of dye mass biosorbed in the column (m_{ad}) is calculated from the area above the breakthrough curve (outlet dye concentration vs. time) multiplied by the flow rate (F). Dividing the dye mass (m_{ad}) by the biosorbent mass (M) leads to the uptake capacity (Q) of the biomass.

The other column parameters were calculated as described below [22]:

- Overall sorption zone:

$$\Delta t = t_e - t_b \tag{1}$$

- Length of the mass transfer zone:

$$L_m = L\left(1 - \frac{t_b}{t_e}\right) \tag{2}$$

- Effluent volume:

$$V_{eff} = Ft_e \tag{3}$$

- Total amount of RB5 sent to column:

$$m_{total} = \frac{C_0 F t_e}{1000} \tag{4}$$

- Total RB5 removal (%):

$$\text{Removal } (\%) = \frac{m_{ad}}{m_{total}} \times 100 \tag{5}$$

The dye mass desorbed (m_d) can be calculated from the area below the elution curve (outlet RB5 concentration vs. time) multiplied by the flow rate. The elution efficiency can be calculated from

$$E\ (\%) = \frac{m_d}{m_{ad}} \times 100 \tag{6}$$

3.2. Modeling of column data

Breakthrough curves obtained at different bed heights (L), flow rates (F) and initial solute concentrations (C_0) were analyzed using three mathematical models; the Thomas, Yoon–Nelson and modified dose–response models, as represented below:

- Thomas model:

$$\frac{C_0}{C} = 1 + \exp\left(\frac{k_{TH}}{F}(Q_0 M - C_0 V_{eff})\right) \tag{7}$$

- Yoon–Nelson model:

$$\frac{C}{C_0} = \frac{\exp(k_{YN}t - \tau k_{YN})}{1 + \exp(k_{YN}t - \tau k_{YN})} \tag{8}$$

- Modified dose–response model:

$$\frac{C}{C_0} = 1 - \frac{1}{1 + (C_0 V_{\text{eff}}/Q_0 M)^{a_{\text{mdr}}}} \quad (9)$$

where k_{TH} is the Thomas model rate constant (l/(mg h)), Q_0 the maximum solid-phase concentration of the solute (mg/g), k_{YN} the Yoon–Nelson model rate constant (1/min), τ the time required for 50% sorbate breakthrough (min) and a_{mdr} the modified dose–response model constant. All model parameters were evaluated using a non-linear regression with the Sigma Plot (Version 4.0, SPSS, USA) software.

4. Results and discussion

Preliminary column RB5 biosorption experiments were performed using free cells of *C. glutamicum*. However, the biomass showed a tendency to form dense slurry that blocked the liquid flow; hence, a high pressure developed and the column became inoperable. Therefore, a possible alternate solution, i.e. immobilization of *C. glutamicum*, was employed. The polysulfone-immobilization process yielded a stable and granular bead that was wheatish in color.

4.1. Influence of bed height

Initial experiments were performed to study the effect of bed height on the biosorption of RB5. Fig. 1 represents the breakthrough curves (C/C_0 vs. time) at different bed heights. The overall performance of a flow-through biosorption column is strongly related to the column breakthrough, exhaustion, length and shape of the sorption zone, uptake capacity and removal efficiency; and thus, these parameters were evaluated and are presented in Table 1. In general, flat breakthrough curves were observed at all bed heights examined. During biosorption using immobilized matrices, intraparticle diffusion usually plays a significant role [11], which is due to the biomass being retained in the interior of the immobilized matrix, so the binding sites are not freely exposed to the solute. Hence, the sorption becomes a multi-step process, with a long time delay occurring between the column breakthrough and exhaustion, resulting in flat breakthrough curves.

Both the breakthrough and exhaustion times increased with increasing bed height, as more binding sites became available for sorption, which also resulted in a broadened sorption zone. As the bed height increased, the steepness of the breakthrough curve decreased; which can also be represented by the slope of the breakthrough curve measured from t_b to t_e (Table 1). The immobilized *C. glutamicum* was observed to possess excellent RB5 biosorption capacity and removal efficiency; 88.9 mg/g and 61.8%, respectively, at a bed height of 10 cm. As expected, a slightly inferior biosorption performance was observed with lower bed heights, because biosorption performance usually depends on the amount of sorbent available for sorption [22].

4.2. Influence of flow rate

Flow rate is an important characteristic affecting the performance of a biosorbent in the continuous mode. The effect of flow rate was studied by fixing the bed height and initial RB5 concentration at 10 cm and 50 mg/l, respectively. The breakthrough profiles are illustrated in Fig. 2, and the column parameters presented in Table 1. Earlier breakthrough and exhaustion were observed at higher flow rates, due to the insufficient solute residence time [23]. The residence time of the solute inside the column is an important parameter in the design of a biosorption packed column procedure. External mass transfer will control the process when the flow rate is very low; conversely, higher flow rates will not be ideal for intraparticle diffusion systems. With high flow rates, not all the solute in the solution will have sufficient time to penetrate to and react with the functional groups, which usually results in a shorter breakthrough time, i.e. improper utilization of the biosorption capacity.

High flow rates also affected the biosorption potential of *C. glutamicum*, with a decrease in RB5 uptake and removal efficiency observed at 1 ml/min. Even though more a shortened sorption zone and relatively steep breakthrough curves were observed at higher flow rates, the biosorption performance was actually better at the lowest flow rate.

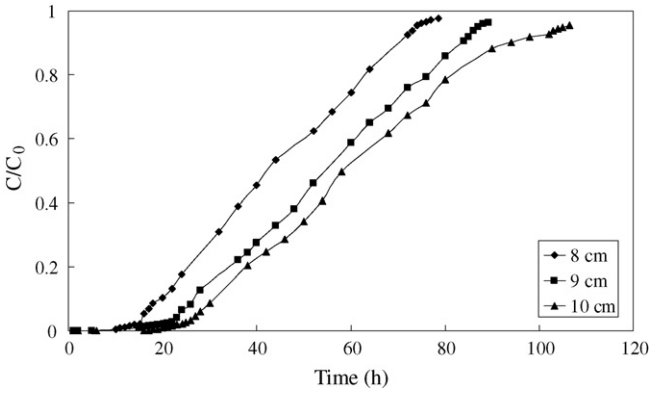

Fig. 1. Experimental breakthrough curves of the removal of RB5 with different bed heights (flow rate = 0.5 ml/min; initial RB5 concentration = 50 mg/l).

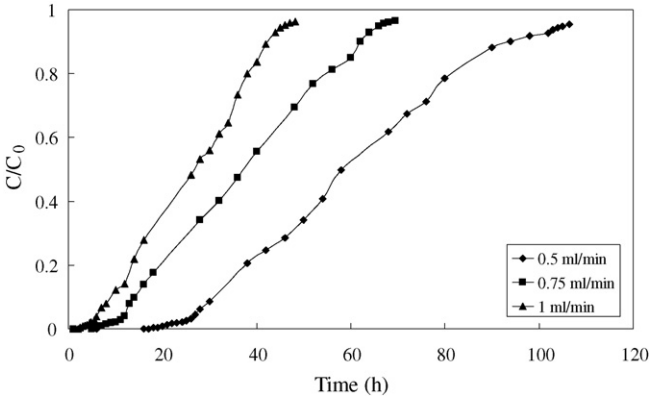

Fig. 2. The effect of flow rate on the biosorption of RB5 by polysulfone-immobilized *C. glutamicum* (bed height = 10 cm; initial RB5 concentration = 50 mg/l).

Table 1
Column data and parameters obtained with different bed heights, flow rates and initial dye concentrations

Z (cm)	F (ml/min)	C_0 (mg/l)	t_b (h)	t_e (h)	Uptake (mg/g dry beads)	dc/dt (mg/(l h))	RB5 removal (%)
8	0.5	50	15.1	78.6	82.9	0.768	60.7
9	0.5	50	20.3	89.2	83.4	0.708	61.1
10	0.5	50	24.2	106.5	88.9	0.597	61.8
10	0.75	50	10.1	69.4	84.3	0.823	58.9
10	1	50	4.8	48.2	73.1	1.148	55.1
10	0.5	75	16.4	87.2	94.2	1.096	52.3
10	0.5	100	9.1	71.9	103.2	1.549	52.1

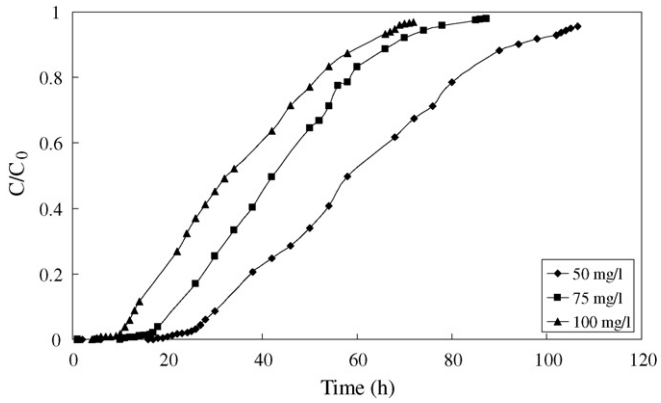

Fig. 3. The effects of initial dye concentration on the biosorption of RB5 by polysulfone-immobilized *C. glutamicum* (bed height = 10 cm; flow rate = 0.5 ml/min).

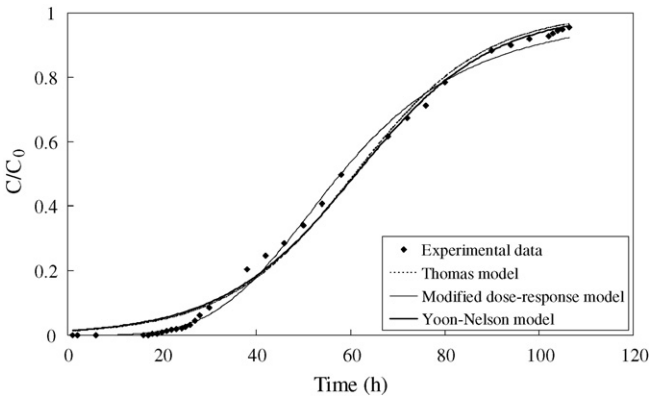

Fig. 4. Application of the Thomas, modified dose–response and Yoon–Nelson models to the experimental data with a bed height, flow rate and initial RB5 concentration of 10 cm, 0.5 ml/min and 50 mg/l, respectively.

4.3. Influence of initial dye concentration

In the subsequent experiments using a column packed with immobilized *C. glutamicum*, the initial RB5 concentration was varied from 50 to 100 mg/l; while the bed height and flow rate were kept constant, at 10 cm and 0.5 ml/min, respectively. As observed in the breakthrough curves in Fig. 3, the breakthrough and exhaustion times decreased with increasing initial dye concentration, as a high concentration leads to rapid saturation of the biomass and earlier column stoppage. This in turn will lead to steeper breakthrough curves, as obvious in Table 1. Maximum RB5 uptakes were obtained with the highest initial dye concentration. The driving force for biosorption is the concentration difference between the dye on the biosorbent and that in the solution [24]. Thus, the high driving force due to a high RB5 concentration resulted in better dye uptake. However, it should be noted that the dye removal efficiency decreased with increasing initial dye concentration. This is because at low solute concentrations, the functional groups in the biosorbent may be sufficient to accommodate all the solute molecules, i.e. fractional sorption becomes independent of the initial solute concentration. With increasing solute concentration, the number of available sites for sorption become fewer compared to the moles of dye present; hence, the percentage of RB5 removed is dependent on the initial dye concentration [25].

4.4. Mathematical modeling of breakthrough curves

The breakthrough curves obtained with different bed heights, flow rates and initial dye concentrations were described using the Thomas, Yoon–Nelson and modified dose–response models. The Thomas model is one of most general and widely used sorption models for describing column breakthrough data. This model assumes Langmuir kinetics of sorption–desorption and no axial dispersion, and is derived with the assumption that the sorption is the rate driving force and obeys second-order reversible reaction kinetics [18]. The Thomas model constants (Q_0 and k_{TH}), determined under different conditions, along with the correlation coefficients, are presented in Table 2. The rate constant (k_{TH}), which characterizes the rate of solute transfer from the liquid to the solid phase, increased with decreasing bed height, increasing flow rate and decreasing initial dye concentration. Conversely, the maximum solid-phase of the solute (Q_0) exhibited a reverse trend (Table 2). This result was as expected for immobilization systems, since high rate constants will usually under utilize the biosorption potential of the biomass due to the possible involvement of intraparticle resistance. Similar results were obtained during the biosorption of phenol by immobilized activated sludge [18]. From Tables 1 and 2, it should also be noted that the Thomas model reasonably predicted the RB5 uptake values and fitted the experimental data well, with high correlation coefficients.

Although very high correlation coefficients were obtained using the Thomas model, it has a fixed value when the experimental time or bed volume is zero (Eq. (7)); which is contrary to real conditions. Yan et al. [26] proposed a modified dose–response model, which minimizes the error that results from use of the Thomas model, especially with lower and higher breakthrough curve times. The model constant (a_{mdr}) increases with increasing bed height, decreasing flow rate and decreasing initial dye concentration (Table 2). Even though the maximum solid-phase of the solute (Q_0) predicted by the modified dose–response model exhibited the same trend predicted by the Thomas model, the predicted values do not coincide exactly with the experimental uptake values.

The Yoon–Nelson model is based on the assumption that the rate of decrease in the probability of adsorption for each sorbate molecule is proportional to the probability of sorbate sorption and the probability of sorbate breakthrough on the sorbent [18]. As shown in Table 2, the 50% breakthrough time (τ) increased and the rate constant (k_{YN}) decreased with increasing bed height, decreasing flow rate and decreasing initial dye concentration. The time required for 50% sorbate breakthrough with the Yoon–Nelson

Table 2
Thomas, Yoon–Nelson and modified dose–response model parameters with different bed heights, flow rates and initial dye concentrations

Z (cm)	F (ml/min)	C_0 (mg/l)	Thomas model			Yoon–Nelson model			Modified dose–response model		
			k_{TH}	Q_0	R^2	k_{YN}	τ	R^2	a_{mdr}	Q_0	R^2
8	0.5	50	0.0018	81.5	0.963	0.088	44.1	0.964	3.58	69.1	0.929
9	0.5	50	0.0016	84.4	0.988	0.079	55.1	0.985	4.05	78.6	0.934
10	0.5	50	0.0015	87.9	0.989	0.073	61.0	0.981	4.07	79.8	0.989
10	0.75	50	0.0019	83.1	0.979	0.095	37.9	0.979	3.29	72.0	0.892
10	1	50	0.0026	74.9	0.921	0.130	27.2	0.923	2.97	67.4	0.781
10	0.5	75	0.0013	90.1	0.992	0.098	43.6	0.991	3.99	85.3	0.978
10	0.5	100	0.0010	101.7	0.981	0.101	33.2	0.978	3.05	87.2	0.931

Table 3
Sorption and elution process parameters for three sorption–desorption cycles

Cycle no.	Uptake (mg/g dry beads)	t_b (h)	t_e (h)	dc/dt (mg/(l h))	L (cm)	L_m (cm)	V_{eff} (l)	RB5 removal (%)	Time for elution (h)	Elution efficiency (%)	Concentration factor
1	88.9	24.2	106.5	0.597	10	7.73	3.2	60.7	5.3	99.6	10.1
2	86.1	22.8	109.3	0.569	10	7.91	3.3	57.3	5.5	99.2	9.9
3	83.8	21.4	111.4	0.565	9.9	8.00	3.3	54.8	5.0	99.5	11.1

model agreed very well with the experimental data. Also, the model predicted the breakthrough curves very well, with high correlation coefficients. Typical examples of the breakthrough curves predicted for the three models examined are shown in Fig. 4.

4.5. Regeneration of biosorbent

The regeneration and subsequent reuse of a biosorbent is crucial for industrial applications for the reduction of the process costs, the continuous dependency of the process on biosorbent and for the possible recovery of dye molecules. For this purpose, it is desirable to desorb the sorbed dye and regenerate the biosorbent material for another cycle. The column was initially packed with 4.8 g (wet weight)/1.1 g (dry weight) of immobilized beads, yielding an initial bed height and volume of 10 cm and 7.85 ml, respectively. The flow rate and initial dye concentration were fixed at 0.5 ml/min and 50 mg/l, respectively. Table 3 summarizes the breakthrough time, exhaustion time and RB5 uptake for all three cycles examined.

The important function of immobilization in a biosorption process is to allow the liquid to flow through the bed, with minimum resistance, and enough rigidity to withstand the extreme conditions during regeneration process [11]. Polysulfone, known for its stability under extreme acidic and alkaline conditions, performed well in aiding *C. glutamicum* to biosorb RB5 in repeated cycles. With progressive, the sorption zone broadened and breakthrough curve flattened (Fig. 5) as a result of the decreased breakthrough and increased exhaustion times. This behavior may primarily have been due to the gradual deterioration of the biosorbent due to repeated usage [27]. The actual height of the bed remained almost constant during the three cycles, with a weight loss of less than 5.6% by the end of the third cycle. The overall performance of the biosorbent in all three cycles was very satisfactory as both very high dye uptake and removal efficiency were observed. The column bed exhibited capacities of over 83.8 mg/g dry beads and removal efficiencies over 54.8%, in all three cycles. When comparing the important column parameters (t_b, L, uptake and removal efficiency), their decreases were all less than 11.5% by the end of the third cycle, indicating the consistently good performance of the polysulfone-immobilized *C. glutamicum* bed.

The minimum bed length (L_m) required to obtain the breakthrough at $t = 0$ (also called critical bed length) uniformly increased with progressive cycles, indicating the broadened mass transfer zone. Since a uniform decrease in sorption performance was observed with progressive cycles, the life of the biosorbent can be predicted based on the important column parameters. For this purpose, three parameters were taken into consideration, including the breakthrough time, column uptake and critical bed length. The following forms of linear regression can be used:

$$t_b = t_{b,i} + k_b n \quad (10)$$

$$Q = Q_i + k_Q n \quad (11)$$

$$L_m = L_{m,i} + k_L n \quad (12)$$

where $t_{b,i}$, Q_i and $L_{m,i}$ are the initial breakthrough time, column uptake and critical bed length, respectively; k_b, k_Q and k_L represent life factors corresponding to the breakthrough time, uptake and critical bed length, respectively; and n represents the cycle number.

From the plot of t_b versus n (Fig. 6), $t_{b,i}$ and k_b were found to be 25.6 h and 1.4 h/cycle, respectively. Thus, the biosorbent bed can be predicted to have sufficient capacity to avoid the breakthrough at time $t = 0$ for up to 18 cycles. From the plot of Q versus n, the expression $Q = 91.38 - 2.564n$ was also formulated. From this expression, it can be estimated that the bed would be completely exhausted (zero uptake) after 35 cycles. A value of 7.6 cm was determined for $L_{m,i}$, giving $k_L = 0.135$ cm/cycle; implying that breakthrough would appear at time $t = 0$ after 18 cycles, which coincides with the result of Eq. (10). The correlation coefficients for all plots were greater than 0.96. Thus, it can be generalized that the sorption zone would reach the top of the bed after 18 cycles and column bed would be completely exhausted after 35 cycles.

The elution curves obtained during three regeneration cycles are presented in Fig. 7. Since biosorption occurred under acidic conditions, it is logical to use an alkaline elutant for the desorption process. Hence, 0.1 M NaOH was employed as the agent for elution of RB5 from the dye-loaded polysulfone beads. The flow rate was maintained at 1 ml/min to avoid over contact of the elutant with the biosorbent. The elution curves observed in all the cycles exhibited a similar trend; a sharp increase at the beginning, followed

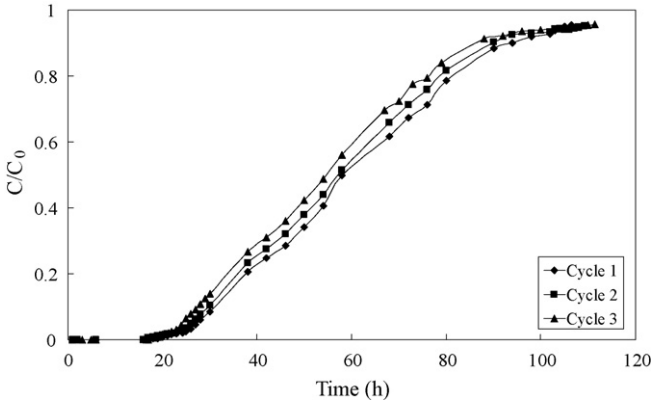

Fig. 5. Experimental sorption breakthrough curves during three regeneration cycles (bed height = 10 cm; flow rate = 0.5 ml/min; initial RB5 concentration = 50 mg/l).

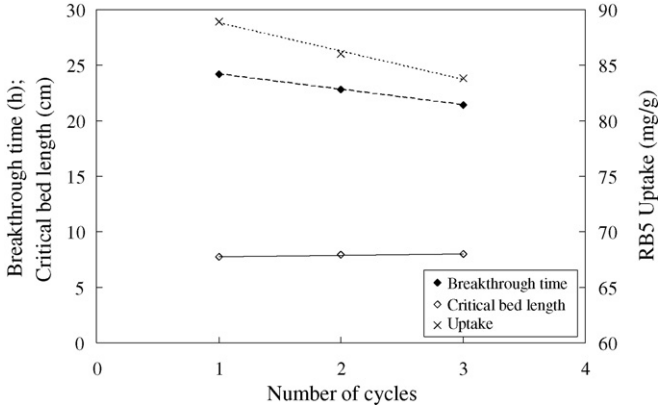

Fig. 6. Linear plots of breakthrough time and RB5 uptake and critical bed length with respect to the number of cycles.

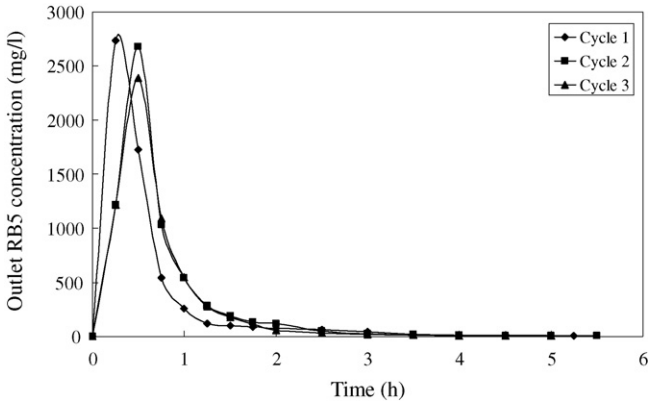

Fig. 7. Column elution curves during three regeneration cycles (elutant = 0.1 M NaOH; flow rate = 1 ml/min).

by a gradual decrease. The elutant performed very well, exhibiting elution efficiencies greater than 99.2% (Table 3). The elution process was carried out for an average of 5.3 h, compared to 109 h for the sorption process, which resulted in highly concentrated dye solutions in only a small volume of elutant. For instance, in cycle 1 at t = 15 min, the effluent RB5 concentration was 2734 mg/l. The concentration factor [28], which can be used to assess the overall success of the biosorption process, can be defined as the ratio of the volume of effluent treated (in sorption process) to that of elutant used (in desorption process). The overall concentration factor for all three cycles was 10.4.

5. Conclusions

This study investigated the potential use of *C. glutamicum* for the removal of RB5 from aqueous solution in an up-flow packed column. As such, the free cells of *C. glutamicum* cannot be employed in packed column due to the increased pressure difference and difficulty in solid–liquid separation. Realizing this, a possible alternate solution, i.e. immobilization, was employed in the present study. The free cells of *C. glutamicum* were immobilized in a polysulfone matrix, which was subsequently used in a column for the continuous biosorption of RB5 from aqueous solution. The crucial column parameters affecting the dye biosorption, such as the bed height, flow rate and initial dye concentration were studied. The results suggest the highest bed height (10 cm), lowest flow rate (0.5 ml/min) and lowest initial dye concentration (50 mg/l) provide in the most favorable outcome. The breakthrough curves obtained under different conditions were described using the Thomas, modified dose–response and Yoon–Nelson models. Regeneration experiments revealed the potential for reuse of the biosorbent using 0.1 M NaOH as the eluting agent. Even though a slight decrease in biosorption performance was observed in subsequent cycles, the polysulfone beads performed excellently in biosorption, maintaining consistently high RB5 uptakes; greater than 83.8 mg/g. Thus, polysulfone-immobilized *C. glutamicum* can be concluded to be an excellent and practical biosorbent for RB5. The application of the findings of this study could be useful in the remediation of reactive dye-bearing industrial effluents.

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