Adsorption of Vancomycin on Amberlite XAD-16 in a Packed Bed Column

B. Borin[a] and A. Pavko[b,*]

[a] Lek Pharmaceuticals d.d., a Sandoz Company, Kolodvorska 27, SI-1234 Mengeš, Slovenia

[b] Faculty of Chemistry and Chemical Technology, University of Ljubljana, Aškerčeva 5, SI-1000 Ljubljana, Slovenia

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Dedicated to the memory of Professor Dr. Valentin Koloini

Vancomycin is a glycopeptide used in the prophylaxis and treatment of bacterial infections. It is obtained with fermentation of bacteria called *Amycolatopsis orientalis*. After fermentation, it is purified of coloured additions and inorganic salts by adsorption to a polymeric adsorbent Amberlite XAD-16. In this work, equilibrium adsorption studies to determine the adsorption capacity were done first. The experimental data fitted best with Langmuir isotherm. Maximum capacity was q_0 = 53.76 kg m^{-3} resin at pH 7. Next, adsorption was studied in a laboratory packed bed adsorption column. A simple mathematical model taking into account axial dispersion was applied to describe the dynamics of the process with breakthrough curves. Experimental and predicted results were compared, and some parametric sensitivity analysis was made to better understand the process for the purpose of scale up.

Key words:
Adsorption, packed bed column, vancomycin

Introduction

Adsorption is a surface phenomena, where components of a gas or liquid are concentrated on the surface of the solid particles. Four types of adsorption are known: exchange, physical, chemical and non-specific. It is a unit operation, widely used in isolation and purification processes of chemical and related industries as well as in environmental technology. Adsorption operation, which consists of three main stages like adsorption, desorption or elution and regeneration of adsorbent, is usually performed in columns with fixed or fluidized beds of adsorbent or in stirred tank reactors. Very often, polymeric resins are used for this purpose. Adsorption is an equilibrium process, which determines the extent to which the material can be adsorbed onto a particular surface. These data are represented in the form of adsorption isotherms, which play a crucial role in the performance of an adsorption system. Several types of these isotherms have been developed, however Langmuir or Freundlich isotherms are the most frequently used to describe this process.[1–4]

Langmuir isotherm is presented with the following equation:

$$q = q_0 \cdot \frac{b \cdot \gamma}{1 + b \cdot \gamma} \qquad (1)$$

where q is equilibrium biosorption capacity of vancomycin, γ is concentration of vancomycin, q_0 is monomolecular capacity and presents the maximum quantity of adsorbed substance per unit of adsorbent mass. Parameter b presents an equilibrium constant, connected with affinity to bonding places of adsorbent.[2–4]

Performance of adsorbers is usually controlled by adsorption rate, which depends on mass transfer processes inside and outside of adsorbent particles. In fixed bed adsorbers, the quality of liquid flow through the porous bed of particles also affects this operation. It is an unsteady-state process, where effluent concentration is a function of time and is presented in a form of breakthrough curve.[1] To describe this process, various mathematical models were developed and confirmed with experimental data which were presented in numerous articles, dealing mostly with metal ions. In contrast, despite the production and consequently isolation of a large number of pharmaceutical compounds in the form of larger molecules, articles dedicated to their recovery with adsorption are very rare. Only papers dealing with in situ recovery of penicillin acylase,[5] recovery of Vitamin B_{12} on non-ionic polymeric adsorbents,[6] modeling of adsorption of a bacterial lipase,[7] and recovery of cephalosporin C on polymeric adsorbent[8] were found in the available literature.

The advection dispersion reaction (ADR) model was applied in the study of vancomycin ad-

* Corresponding author: E-mail: saso.pavko@fkkt.uni-lj.si

sorption presented here. This model was already studied by many other authors.[9–12]

This simple model is based on material balance equations in the liquid phase and solute transport and its adsorption by the solid phase. The following equation can be solved for single solute and one dimension:

$$\frac{\partial \gamma}{\partial t} = D_{ax} \cdot \frac{\partial^2 \gamma}{\partial z^2} - u_z \cdot \frac{\partial \gamma}{\partial z} - \frac{\rho_s(1-\varepsilon)}{\varepsilon} \cdot \left(\frac{\partial q}{\partial t}\right) \quad (2)$$

On the right side of this equation, the first two parts describe diffusive motion and bulk motion of the whole solution while the third term presents adsorption process. Solution of equation by time t and coordinate z describes the performance and process dynamics in a packed bed column. Graphical solution presents breakthrough curve, which is profile of concentration in the column outlet as the function of time.

An important part in this equation is the adsorption term, in our case presented by the derivative of the Langmuir equilibrium equation, which finally gives the model equation:

$$\frac{\partial \gamma}{\partial t} = D_{ax} \cdot \frac{\partial^2 \gamma}{\partial z^2} - u_z \cdot \frac{\partial \gamma}{\partial z} - \frac{\rho_s(1-\varepsilon)}{\varepsilon} \cdot \left(\frac{q_0 \cdot b}{(1+b\cdot\gamma)^2}\right) \cdot \frac{\partial \gamma}{\partial t} \quad (3)$$

In order to solve this equation, the following parameters are needed:

– the sorptive behavior of polymeric adsorbent, in our case Langmuir parameters q_0 and b,

– the density of biosorbent material (ρ_s)

– porosity of packed bed (ε)

– operating conditions like volumetric flow rate (F) and input solute concentration (γ_0)

– the column geometric characteristics to determine the interstitial velocity $u_z = F/\varepsilon A$, where A is the cross-section area of the void column,

– deviation of ideal plug flow, expressed with axial dispersion coefficient D_{ax}

The following boundary conditions were taken into account:

$$t = 0, \quad z = 0; \quad \gamma = \gamma_0,$$

$$z = L; \quad d\gamma/dz = 0$$

Initial condition:

$$\gamma = 0 \qquad 0 \le z \le L, \; t = 0$$

Boundary conditions:

$$\left.\frac{\partial \gamma}{\partial z}\right|_{z=L} = 0 \qquad \text{at } 0 < t < t_F$$

$$D_{ax} \cdot \left.\frac{\partial \gamma}{\partial z}\right|_{z=0} = v \cdot (\gamma_0 - \gamma) \qquad \text{at } 0 < t < t_F$$

where t_F is the calculation time. A detailed review of the solutions of this ADR model is available elsewhere.[3,11]

Vancomycin is a glycopeptide used in the prophylaxis and treatment of bacterial infections.[13] It is a fermentation product, recovered from cultivation broth after fermentation by adsorption to a polymeric resin. The aim of this investigation was to determine the adsorption capacity on polymer resin Amberlite XAD 16 and to describe the dynamics of the process in a laboratory packed bed column for the further scale up of this operation.

Materials and methods

Determination of adsorption capacity

Experiments for the determination of isotherms to determine the adsorption capacity were performed with 25 mL batches of pure vancomycin solutions in 250 mL Erlenmeyer flasks with magnetic stirrers at room temperature. 10 mL batches of regenerated polymer resin Amberlite XAD 16 (Rohm&Haas Company, USA)[14] with particle density ρ_s = 1.02 kg m^{-3} and size d_p = 0.635 mm were mixed with the vancomycin solutions having the following initial concentrations: 2, 5, 10, 15, 20, 25 and 30 kg m^{-3}. Mixing during experiments was adjusted to 100 rpm and the equilibrium was reached in less than 30 minutes. Experiments were performed at pH 7.0 ± 0.2. HPLC analysis (1200 SL, Agilent Technologies, USA, wavelength 275 nm) was used to determine the vancomycin concentration.

Adsorption in a laboratory packed bed column

The experiments were performed in a 0.45 m long glass column with 0.025 m diameter. The column was filled with a regenerated polymer resin so that 0.18 m high bed was obtained. The bed porosity ε = 0.41 was determined in a separate experiment as follows: resin was first soaked in demineralized water to remove the solvents used during regeneration, drained to remove excess water and surface dried. The 100 mL graduated measuring cylinder was filled with resin to the mark and then filled with water to the same mark. Porosity was then cal-

culated as the quotient of the measured volume of added water and the total volume.

The column was connected to pulse pump (Gamma/4-1, Prominent GmbH, Germany) to feed the column with the solution of γ_0 = 11.5 kg m^{-3} vancomycin and pH 7. The level of liquid above the resin was kept steady to achieve the liquid flow rate $2 \cdot 10^{-8}$ m^3 s^{-1}. The outlet of the column was connected to a fraction collector (Fraction Collector 684, Büchi Labortechnik AG, Switzerland) where every 25 minutes a fraction was collected. All data and operation conditions are given in Table 1.

Model calculations

Eq. 3 was converted into difference form and solved with a finite differences method using Wolfram Research Company software package Mathematica 5.[15] Concentration of vancomycin in radial direction was assumed to be equal, so the equation was solved only in longitudinal direction. The column was divided into 100 parts of equal length. After each calculation of concentration for a part of the column, the next step for time evaluation was made so that time increments of 10 s were taken into account. The calculation of breakthrough curve for column was finished after t_F = 65000 seconds.

Parametric sensitivity analysis was made for breakthrough time, which means the time when outlet concentration reaches 5 % value of inlet concentration and presents the beginning of breakthrough curve. It is shown graphically where relative change is presented against the relative change of the parameter under investigation.[11]

Results and discussion

The results of equilibrium adsorption experiments at pH = 7 to determine the adsorption capacity are presented in Fig. 1. Experimental data were better fitted with Langmuir equation (R^2 = 0.99) than with Freundlich equation (R^2 = 0.95). The Langmuir parameters, maximum adsorption capacity q_0 = 53.76 kg m^{-3} resin (0.043 kg kg^{-1} resin) and equilibrium constant b = 6.64 m^3 kg^{-1} were used in model calculations.

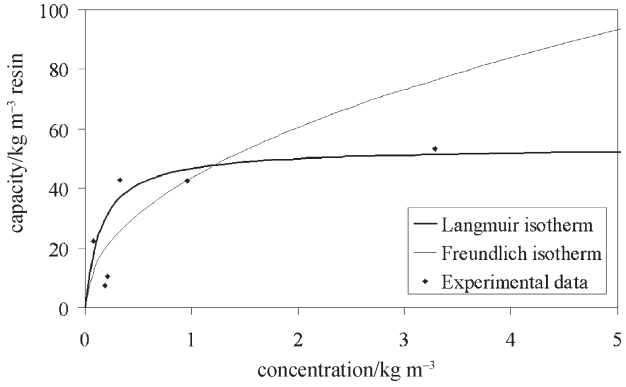

Fig. 1 – *Results from the equilibrium experiments correlated with Langmuir and Freundlich isotherm*

Experimental results from the column in the form of breakthrough curve together with the predicted results are presented in the Fig. 2. The parameters for model calculations are presented in Table 1. In addition to this figure, the results of parametric sensitivity analysis are presented in Fig. 3. During this analysis, only one parameter was varied at a time, the others being set as estimated values as presented in Table 1.

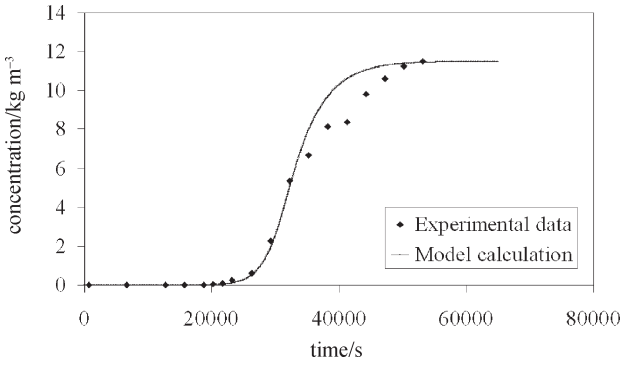

Fig. 2 – *Experimental results and simulated breakthrough curve in a packed bed column for the data from Table 1*

Table 1 – *Data for the model calculations*

Parameter	Unit	Value
q_0	kg m^{-3}	53.76
b	m^3 kg^{-1}	6.64
ρ_s	kg m^{-3}	$1.02 \cdot 10^3$
γ_0	kg m^{-3}	11.5
F	m^3 s^{-1}	$2 \cdot 10^{-8}$
L	m	0.18
A	m^2	0.00049
ε	/	0.41
D_{ax}	m^2 s^{-1}	$4.9 \cdot 10^{-5}$

The parameter sensitivity analysis in Fig. 3 shows that the maximum biosorption capacity has the expected substantial effect on the breakthrough time. An increase of the maximum biosorption capacity proportionally increases the bed capacity and consequently prolongs the breakthrough time. The material density (see eq. 3) has a similar influence on the position of a breakthrough curve. The in-

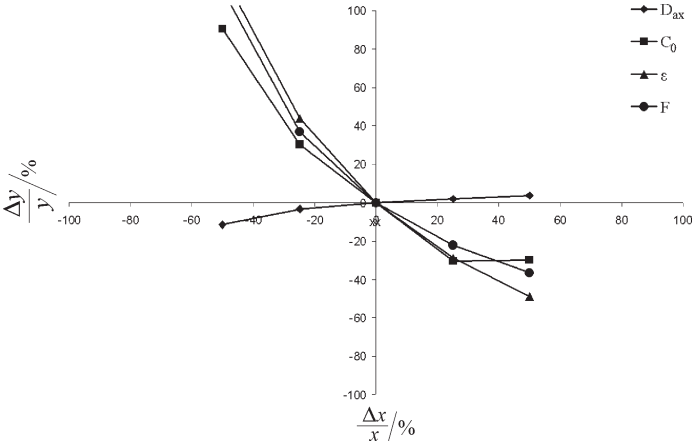

Fig. 3 – *Parametric sensitivity analysis showing the effect of various parameters on breakthrough time in a packed bed column*

crease of material density increases the mass of the adsorption material of a given volume and therefore moves the curve to the right towards higher breakthrough times. The initial vancomycin concentration and the breakthrough time are interrelated through the rate of adsorption. At the same adsorption time, a higher initial concentration causes an earlier breakthrough because of the solid bed material inability to adsorb more solute in the same time. The effect of liquid flow rate is also expected. Its increase reduces the residence time in the column and consequently causes earlier breakthrough.

The mathematical model is very sensitive to the bed porosity. The literature gives the porosity of packed bed columns between 0.35 and 0.5[16] while the measured porosity in this work was $\varepsilon = 0.41$ which is in very good agreement with the literature data. Furthermore, the experimental results and the predicted results of breakthrough curve with $\varepsilon = 0.41$ are in excellent agreement (see Fig. 2). Increase of bed porosity causes a decrease in breakthrough time. On one hand, the increase of bed porosity at a given flow rate F decreases the liquid velocity and consequently axial dispersion coefficient as well as the mass of biosorbent material, whilst on the other hand increases the residence time in the column. All this explains the reason for earlier breakthrough. This trend of results is consistent with the results from the literature.[3,11]

Axial dispersion coefficient was estimated with correlation equation by Langer *et al.* (1978)[16] as well as using the correlation by Wakao,[3] where the vancomycin molecular diffusivity was estimated by Othmer-Thakar equation.[17] The obtained values are $4.9 \cdot 10^{-5}$ m^2 s^{-1} and $1.2 \cdot 10^{-4}$ m^2 s^{-1} respectively. Reynolds number was estimated to be Re = 0.03.

Better fit with the experimental data was obtained with the value of axial dispersion coefficient $D_{ax} = 4.9 \cdot 10^{-5}$ m^2 s^{-1}. On one hand, the value of D_{ax} for vancomycin is actually only an extrapolated rough estimation by the used equation, since the necessary data for this molecule are not available in the literature. On the other hand, D_{ax} in the simple model used here is actually an overall apparent value, which combines both the characteristics of the flow and the mass transfer resistances in the liquid and solid phases.[11] Change of D_{ax} by only $+/-$ 50 % has a minor effect on breakthrough time compared to other parameters shown in Fig. 3. It is probably due to relatively short packed bed of adsorbent material, low interstitial velocity and high feed concentration used in this work. Additional calculations were made for longer column ($L = 1.0$ m), 1000 times higher flow rate ($F = 2 \cdot 10^{-5}$ m^3 s^{-1}) and lower feed concentration $\gamma_0 = 1.15$ kg m^{-3}. The calculated breakthrough time and the slope of the curve decreased by increasing D_{ax} value, according to the literature data.[11] For example, by setting the values of D_{ax} to be $4.9 \cdot 10^{-8}$ m^2 s^{-1}, $4.9 \cdot 10^{-5}$ m^2 s^{-1} and $4.9 \cdot 10^{-3}$ m^2 s^{-1}, the calculated breakthrough times were 34.3 h, 12.9 h and 3.3 h, respectively.

Conclusions

Despite the widely used adsorption process as a unit operation in chemical and related industries, articles dedicated to the recovery of larger molecules such as antibiotics with adsorption are very rare. In our work, it was found that the adsorption of vancomycin can be successfully applied on polymeric resin Amberlite XAD 16. It can be well described with a Langmuir isotherm. Our experimental results of breakthrough curves show that the adsorption process as a downstream operation can be done in a packed bed column. These results were compared with predicted results, using a simple mathematical model. Axial dispersion coefficient as an important model fitting parameter shows a possible influence of internal and external mass transfer as well as flow and packed bed characteristics on the adsorption process. As expected, the breakthrough curves are also sensitive to adsorbent characteristics and other operating parameters such as bed porosity, liquid flow rate and initial concentration. The results present a good basis for further investigation and optimization in a pilot plant column and further application on the industrial scale.

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List of symbols

A – column cross-section area, m^2
b – Langmuir coefficient, $m^3\ kg^{-1}$
γ – concentration of vancomycin, $kg\ m^{-3}$
γ^* – equilibrium concentration of vancomycin, $kg\ m^{-3}$
γ_0 – feed concentration of vancomycin, $kg\ m^{-3}$
D_{ax} – axial dispersion coefficient, $m^2\ s^{-1}$
F – liquid flow rate, $m^3\ s^{-1}$
L – bed length, m
q – equilibrium biosorption capacity of vancomycin, $kg\ m^{-3}$
q_0 – maximum biosorption capacity of vancomycin, $kg\ m^{-3}$
t – time, s
t_F – model calculation time, s
u_z – liquid phase interstitial velocity, $m\ s^{-1}$
z – longitudinal coordinate, m
ρ_s – biosorbent material density, $kg\ m^{-3}$
ε – bed porosity, –

References

1. *Bathen, D., Breitbach, M.,* Adsorptionstechnik, Spinger, Berlin, 2001.
2. *Doran, P. M.,* Bioprocess Engineering Principles, Academic Press Ltd, London, 1995, pp 234-40.
3. *Ruthven, D. M.,* Principles of Adsorption and Adsorption Processes, John Wiley & Sons New York, 1984.
4. *Hauffe, K.,* in *Morrison, R.,* Adsorption: eine Einführung in die Probleme der Adsorption, Walter de Gruyter, Berlin, 1974.
5. *Fonseca, L. P., Cabral, J. M. S.,* J. Chem. Technol. Biotechnol. **77** (2002) 1176.
6. *Ramos, A. M., Otero, M., Rodrigues, A. E.,* Separation and Purification Technology **38** (2004) 85.
7. *Millitzer, M., Wenzig, E., Peukert, W.,* Biotechnology and Bioengineering **92** (2005) 789.
8. *Mishra, P., Srivastava, P., Mishra, P. K., Kundu, S.,* Indian Journal of Chemical Technology **14** (2007) 592.
9. *Farthing, M. W., Kees, C. E., Russell, T. F., Miller, C. T.,* Adv. Water Res. **29** (2006) 657.
10. *Ghorai, S., Pan, K. K.,* Chem. Eng. J. **98** (2004) 165.
11. *Hatzikioseyian, A., Tsezos, M., Mavituna, F.,* Hydrometallurgy **59** (2001) 395.
12. *Nofziger, D. L., Wu, J.* (2003) *Convective-Dispersive Transport with Steady-State Water Flow.* [web document]. Accessed on September 7th, 2009 on http://soilphysics.okstate.edu/software/cde/document.html.
13. *Bunch, A.* in *Baines, A.* Vancomycin – a vital antibiotic [web document]. Accessed on September 7th, 2009 on http://www.chm.bris.ac.uk/motm/vancomycin/vanc.htm.
14. *Rohm and Haas Amberlite XAD 16 Industrial Grade Polymeric Adsorbent. Product data Sheet.* [web document]. Accessed on September 13th, 2009 on http://i01.yizimg.com/upload/140240/ /20069261323102503012 58.pdf
15. Wolfram Research. *Software package Mathematica 5.0.*
16. *Perry, R. H., Green, D. W.,* Perry's Chemical Engineers' Handbook on CD-ROM, 16-21, McGraw-Hill Companies, New York, 1999.
17. *Sherwood, T. K., Pigford, R. L., Wilke, C. R.,* Mass Transfer, McGraw-Hill, New York, 1975, pp 29.