On the approximate analytical solution of a problem of optimization in the field of immobilized enzymes

by F. XAVIER MALCATA

Escola Superior de Biotecnologia, Universidade Catolica Portuguesa,
Rua Dr Antonio Bernardino de Almeida, 4200 Porto, Portugal

The problem of optimizing the thickness of a microporous slab containing an immobilized enzyme is addressed, using an economic criterion as the objective function. The steady-state material balance to the substrate as transported by diffusion and depleted by a biochemical reaction following classical Michaëlis–Menten kinetics within the pellet is obtained. Taking advantage of a number of algebraic manipulations and mathematical artefacts, one is able to solve the resulting second-order, non-linear differential equation by an analytical method, provided that an upper error bound for the solution in the order of 5 per cent is acceptable. The validity of the approximation is tested, and useful applications are reported.

1. Introduction

Enzymes are proteins with enhanced catalytic properties due to their power of specific activation [1]. As proteins, enzymes consist of a linear chain of amino acid residues bonded via peptide linkages. As catalysts, enzymes affect the rate of biochemical reactions without themselves appearing as either reactants or products of the global reaction, and without being consumed within reasonable time scales. Being highly specific, enzymes act only upon some bonds of some substrates, thus accelerating chemical reactions without formation of by-products. Furthermore, enzymes are able to function in dilute aqueous solutions under very mild conditions of temperature, pH and ionic strength [2]. These characteristics have emphasized the industrial use of enzymes to bring about a number of relevant chemical reactions within the food, pharmaceutical, and cleaning fields.

Immobilization of an enzyme on or within an insoluble solid matrix means that the enzyme has been confined or localized so that it can be re-used continuously [3]. There are several reasons why immobilization may be desirable: no enzyme is allowed to leave the reactor in the outlet solution (which avoids the need of extra enzyme makeup, or of enzyme removal from the product where it might play the role of an impurity); longer retention of catalytic activity is possible (so the enzyme becomes intrinsically more stable and less sensitive to thermal denaturation); and the control of the chemical process becomes more feasible and accurate (thus eliminating unwanted variability and making it easier to meet the required product specifications). These characteristics make immobilized enzyme technology attractive if a very large throughput is required and/or the enzymes involved are expensive. Three notable examples of industrial processes that employ immobilized enzyme catalysts are the production of high fructose corn syrups from corn starch, the manufacture of L-amino acids by resolution of racemic amino acid mixtures, and the manufacture of semi-synthetic penicillins [3].

Many methods are available for enzyme immobilization. Enzymes are typically immobilized on the internal surfaces of porous supports, or entrapped in matrices through which a substrate can diffuse. In such systems, calculation of the observed rate of substrate disappearance requires evaluation of the concentration profile of substrate within the pellet [3]. Fick's law of diffusion can in principle be applied [4], provided that the effective diffusivity is known. This parameter accounts for the fact that some of the particle cross-section is occupied by solid and hence not available for diffusive transport, and the pore network is complex and entangled so diffusion occurs only in allowed, frequently changing directions [5]. On the other hand, the kinetic equation describing the biochemical reaction occurring within the pellet must be taken into account; this is usually done via the utilization of the classical Michaëlis–Menten model [6], where the order of the reaction on the substrate concentration increases gradually from zero (for high substrate concentrations) up to unity (for low substrate concentrations).

A number of shapes for the pellets where the enzymes can be immobilized are commercially available, the slab-shaped type being particularly easy to manufacture and model. The development of a design methodology for optimal slab thickness in heterogeneous, catalytic biochemical reactors is, then, undoubtedly both of academic interest and commercial significance.

2. Problem formulation

Consider a control volume accounted for by a porous solid supporting an immobilized enzyme, whose pores are completely filled with a liquid solution containing the reacting substrate. A chemical reaction following Michaëlis–Menten kinetics takes place within this porous pellet, with a 1 : 1 stoichiometry [7]. This stoichiometry, coupled to the assumption that the solvent is virtually stagnant, leads to a null molar average velocity, which is characteristic of an equimolar counter diffusional process. The solute becomes available to the active sites inside the pellet via a molar diffusion flux arising from a concentration gradient across the particle. The physical medium is assumed to behave isotropically with respect to the global mass density and to the resistance offered to the molecular transport of solute within the pellet. The resistance to mass transfer of the liquid film surrounding the pellet is considered negligible when compared with the intraparticle resistance. The average pore size is taken as sufficiently low so that the external forced convection past the pellet does not penetrate the pore network, and thus has no appreciable effect on the intraparticle concentration profile [8]. The solution containing the substrate is assumed to behave ideally, so no volume change of mixing occurs. Steady-state conditions of operation have been reached. The process of diffusion and homogeneous reaction can then be described [9–12] by

$$D_{\mathrm{eff}}\nabla^2 C = \frac{v_{\mathrm{m}} C}{K_{\mathrm{m}} + C} \tag{1}$$

where C denotes the concentration of substrate, D_{eff} is the effective diffusivity of substrate within the pellet, v_{m} is the maximum rate, and K_{m} is the Michaëlis–Menten constant. In the case of a constant cross-sectional area, sealed-edge, catalyst slab of half-thickness L in the direction of solute flow (the x-direction), fairly small when compared with the other characteristic dimensions of the slab, the problem becomes virtually unidimensional. Equation (1) can, thus, be simplified to

$$\frac{d^2C}{dx^2}=\frac{v_m}{D_{eff}}\frac{C}{K_m+C} \tag{2}$$

A set of physically meaningful boundary conditions can be written as

$$x=0, \quad \frac{dC}{dx}=0 \tag{3}$$

arising from the symmetry condition at the centre plane of the slab [9], and

$$x=L, \quad C=C_s \tag{4}$$

where the subscript ‘s’ stands for the conditions prevailing at the surface of the slab. An analytical solution of equation (2) coupled to equations (3) and (4) has not been reported to date [13], so a numerical method is required. Since one is dealing with a double boundary value problem, an iterative procedure is in general needed [14]; the integration is then performed through the construction of a concentration profile from the initial point, given by equation (3), as many times as needed for convergence be achieved to the final condition, equation (4). This method may be cumbersome to implement and somewhat susceptible to cumulative errors because a higher-order derivative exists; furthermore, no general conclusions can be drawn out for the problem because each time the parameters are changed the numerical procedure has to be restarted and carried over once more.

An interesting alternative approach can, nonetheless, be devised after a careful inspection of equation (2). In fact, no explict functional dependence on variable x exists; the independent variable can, therefore, be changed from x to C [13, 15] through the definition of the auxiliary dependent variable Φ

$$\Phi\equiv\frac{dC}{dx} \tag{5}$$

Application of the chain rule of differentiation to equation (5) and use of the result in equation (2) eventually gives

$$\Phi\frac{d\Phi}{dC}=\frac{v_m}{D_{eff}}\frac{C}{K_m+C} \tag{6}$$

Equation (3) is now directly replaced by an equivalent boundary condition, namely

$$C=C_c, \quad \Phi=0 \tag{7}$$

where the subscript c denotes the conditions on the symmetry plane of the slab. Adequate integration of equation (6) coupled to equation (7) leads to

$$\Phi^2=\frac{2v_m}{D_{eff}}\left[C-C_c-K_m\ln\left[\frac{K_m+C}{K_m+C_c}\right]\right] \tag{8}$$

A suitable variable normalization is now apparent, according to

$$C^*\equiv 1+\frac{C}{K_m} \tag{9}$$

which leads to the logical definition of the following dimensionless parameter

$$Th \equiv L\left[\frac{v_m}{K_m D_{\mathrm{eff}}}\right]^{1/2} \tag{10}$$

usually known as the Thiele modulus [5]. Using equations (9) and (10) in equation (8), and recalling equations (4) and (5), one finally converts the differential equation to quadratures, namely

$$Th(C_c^*) = \int_{C_c^*}^{C_s^*} \frac{d\xi}{[2[\xi - C_c^* - \ln(\xi/C_c^*)]]^{1/2}} \tag{11}$$

where C_c^* is the current independent variable of interest. An artefact is now possible with respect to the evaluation of the above integral. In fact, the integral term in equation (11) can be partitioned into two integrals, and a new dummy variable of integration can be defined according to

$$\zeta \equiv \ln(\xi/C_c^*) \tag{12}$$

thus leading to

$$Th(C_c^*) = \int_{C_c^*}^{C_s^*} \frac{\left[1 - \dfrac{1}{\xi}\right] d\xi}{[2[\xi - C_c^* - \ln(\xi/C_c^*)]]^{1/2}} + \int_0^{\ln(C_s^*/C_c^*)} \frac{d\zeta}{[2[C_c^*[\exp(\zeta) - 1] - \zeta]]^{1/2}} \tag{13}$$

Expansion of the exponential term in equation (13) as a McLaurin series [15], and truncation after the quadratic term yields

$$Th(C_c^*) \simeq Th_{\mathrm{app}}(C_c^*) \equiv [2[C_s^* - C_c^* - \ln(C_s^*/C_c^*)]]^{1/2} + \int_0^{\ln(C_s^*/C_c^*)} \frac{d\zeta}{[\zeta[C_c^*\zeta + 2(C_c^* - 1)]]^{1/2}} \tag{14}$$

The integral in equation (14) can be obtained as a finite combination of elementary functions, provided that a Chebyshev change of variable of the third kind is performed [16, 17] according to

$$\eta^2 \equiv 1 + \frac{2(C_c^* - 1)}{\zeta} \tag{15}$$

After integration, (14) finally becomes

$$Th_{\mathrm{app}}(C_c^*) = [2[C_s^* - C_c^* - \ln(C_s^*/C_c^*)]]^{1/2} + \frac{1}{\sqrt{C_c^*}} \ln\left[\frac{\left(1 + \dfrac{2(C_c^* - 1)}{C_c^* \ln(C_s^*/C_c^*)}\right)^{1/2} + 1}{\left(1 + \dfrac{2(C_c^* - 1)}{C_c^* \ln(C_s^*/C_c^*)}\right)^{1/2} - 1}\right] \tag{16}$$

An estimate of the error implicit in the foregoing approximation can be obtained by numerically integrating equation (13) by a trapezoidal rule, with a total number of mesh-points equal to $N_{m,tot}$, each one being located in the x-dimension according to the formula

$$x_{m,i} = \delta + [\ln(C_s^*/C_c^*) - \delta]\left[1 - \cos\left[\frac{\pi(i-1)}{2(N_{m,tot}-1)}\right]\right] \tag{17}$$

The fractional error of the approximation can be depicted in Figure 1 for a few levels of parameter C_s^*.

When dealing with a chemical process aimed at the manufacture of a high-value chemical compound via an enzyme-catalyzed biochemical route using fixed-bed reactor technology, the choice of the thickness of the slab can be obtained as a compromise between the market price of the product and the capital cost of the pellet. This statement can be mathematically expressed via

$$V_n = 2AV_pD_{eff}\left[\frac{dC}{dx}\right]_{x=L} t_e - 2ALV_c \tag{18}$$

where V_n is the overall net profit associated with the operation of the reactor over the entire service life of the enzyme, A is the cross-sectional area of the catalyst slab, V_p is the molar market price of the product, V_c is the volumetric market cost of the supported enzyme, and t_e is the service life of the supported enzyme. Equation (18) is equivalent to the dimensionless form

$$V_n^* = Ma[C_s^* - C_c^* - \ln(C_s^*/C_c^*)]^{1/2} - \sqrt{2Th} \tag{19}$$

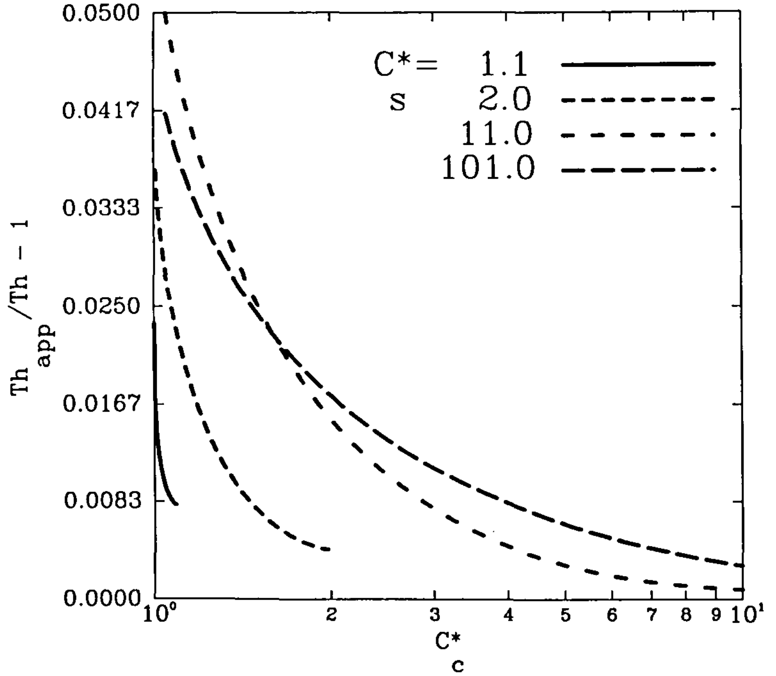

Figure 1. Fractional error of the approximation used to compute Th, $(Th_{app} - Th)/Th$, plotted as a continuous function of C_c^* for various values of C_s^*.

where Ma is a dimensionless economic parameter defined as

$$Ma \equiv \frac{(2D_{\mathrm{eff}})^{1/2} V_{\mathrm{p}} v_{\mathrm{m}} t_{\mathrm{e}}}{V_{\mathrm{c}}} \tag{20}$$

and the dimensionless net profit is obtained from

$$V_{\mathrm{n}}^{*} \equiv \frac{V_{\mathrm{n}} \sqrt{v_{\mathrm{m}}}}{A V_{\mathrm{c}} / (2 D_{\mathrm{eff}} K_{\mathrm{m}})^{1/2}} \tag{21}$$

Differentiating equation (19) with respect to C_{c}^{*}, and setting the outcoming result equal to zero, one obtains

$$\begin{aligned} Ma \simeq\; & 2 + \frac{[C_{\mathrm{s}}^{*} - C_{\mathrm{c,opt}}^{*} - \ln (C_{\mathrm{s}}^{*}/C_{\mathrm{c,opt}}^{*})]^{1/2}}{C_{\mathrm{c,opt}}^{*} - 1}\, x \\ & \times \left\{ \frac{\sqrt{2}}{(C_{\mathrm{c,opt}}^{*})^{1/2}} \ln \left[\frac{\left(1 + \dfrac{2(C_{\mathrm{c,opt}}^{*} - 1)}{C_{\mathrm{c,opt}}^{*} \ln (C_{\mathrm{s}}^{*}/C_{\mathrm{c,opt}}^{*})}\right)^{1/2} + 1}{\left(1 + \dfrac{2(C_{\mathrm{c,opt}}^{*} - 1)}{C_{\mathrm{c,opt}}^{*} \ln (C_{\mathrm{s}}^{*}/C_{\mathrm{c,opt}}^{*})}\right)^{1/2} - 1} \right] \right. \\ & \left. + \frac{2\sqrt{2}(C_{\mathrm{c,opt}}^{*} - 1 + \ln (C_{\mathrm{s}}^{*}/C_{\mathrm{c,opt}}^{*}))}{(C_{\mathrm{c,opt}}^{*} - 1) \ln (C_{\mathrm{s}}^{*}/C_{\mathrm{c,opt}}^{*}) \left(C_{\mathrm{c,opt}}^{*} + \dfrac{2(C_{\mathrm{c,opt}}^{*} - 1)}{\ln (C_{\mathrm{s}}^{*}/C_{\mathrm{c,opt}}^{*})} \right)^{1/2}} \right\} \end{aligned} \tag{22}$$

After the optimum value $C_{\mathrm{c,opt}}^{*}$ for C_{c}^{*} is found via equation (22) for any given Ma, equation (16) can be used to obtain the corresponding value for Th_{opt} as depicted in Figure 2. The calculation of the optimum value of the slab thickness is now apparent from equation (10).

3. Discussion

The traditional approach to the optimal design of the thickness of slab-shaped pellets catalyzing simple biochemical reactions is based on the calculation of an effectiveness factor for the Michaëlis–Menten, rectangular hyperbola-type kinetic expression [18, 19]. A dimensionless parameter containing the Michaëlis–Menten kinetic constant appears to play an important role in such an analysis. The alternative approach reported in this communication replaces that parameter by a dimensionless concentration of substrate at the centre of the slab; this fact arises naturally from the strategy followed during the integration of the differential mass balance to the substrate within the pellet. Although introduced during the analysis in a way similar to a boundary condition, C_{c}^{*} is not truly required to specify any integration constant; C_{c}^{*} turns out to be the actual, physically meaningful, independent variable in the optimization procedure.

The expansion of the exponential function in equation (13) as a power series presents obvious advantages relative to the expansion of the logarithmic function in equation (11). In fact the former possesses an infinite radius of convergence, whereas the latter has a considerably narrow, finite radius of convergence; thus, the expansion actually employed leads to more general and powerful results.

The solution of equation (11) is accomplished at the expense of an approximating function. The integrand of this equation possesses a singularity at the lower limit of

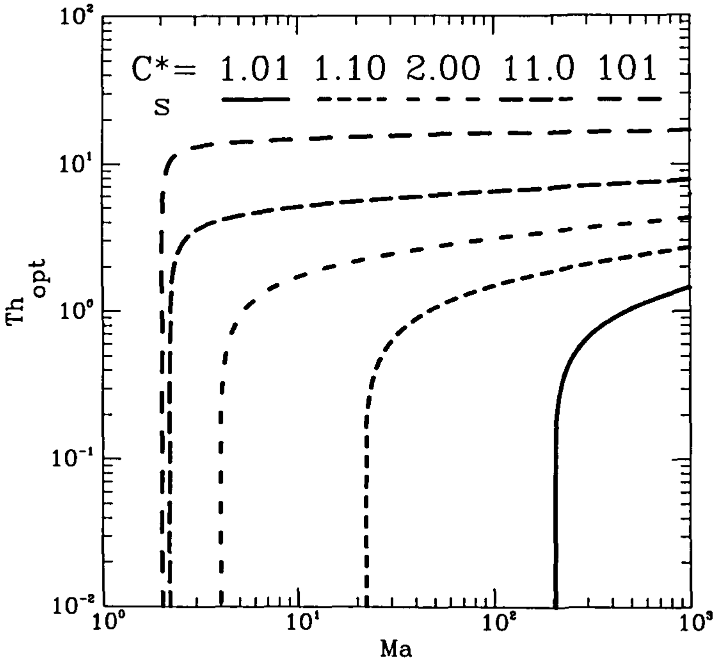

Figure 2. Plot of the values for Th_{app} leading to the optimal economic design Th_{opt} versus the economic parameter Ma for various values of C_s^*.

integration which gives a major contribution to the value of the integral. The use of the approximating function avoids numerical integration, which in this case might be a notoriously error-prone process if no special precautions are taken in order to deal with the aforementioned singularity. The approximation used consists of expanding the exponential function in such a way as to become exact at the lower limit of integration. In order to obtain an estimate of the error associated with the approximation, the numerical integration of equation (13) was accomplished via a trapezoidal rule, using a criteriously defined spacing for the mesh-points as introduced in equation (17). The particular spacing formula employed clusters the points near the lower boundary, where the singularity is more likely to affect the mathematical behaviour of the function. Owing to the finite nature of the hardware available for automatic calculus, integration can only be numerically started at a positive value of δ, the lower boundary condition. The optimal δ to be used, as well as the total number of mesh points required were found by a trial-and-error procedure, aiming at reaching the best compromise between low CPU time and high intrinsic mathematical accuracy; a double-precision methodology was necessary in order to release partly the constraint of additive error during the iterative numerical evaluation of the integral. The percentual error does not exceed the 5 per cent level in the range of physical interest. Larger values for C_c^* lead to a monotonically decreasing fractional error for the approximation. As C_s^* increases, the fractional error tends to become more sluggish with C_c^*, with the worst situation being achieved at an intermediate range of the dimensionless surface concentration (about 11·0). The highest interest lies on the case of having $C_s^* \simeq 2{\cdot}0$, because this corresponds to the physiological level where most enzymes exhibit the best operating characteristics (i.e. $C_s \sim K_m$ leads to the largest efficiency coupled to the highest control feasibility [1]). It is remarkable to note that the approximation resulting from the truncation of the series expansion of the exponential function about zero after the quadratic term always yields consistently positive deviations in the computation of the Thiele modulus. Therefore, a conservative approach results, which makes the approximation developed robust to inadequacies during modelling and/or inaccuracies in the estimation of the parameters relevant for the operation.

As the value of the product of the reaction increases with respect to the cost of the supported enzyme, the optimal concentration of substrate at the centre of the slab decreases, and the corresponding Thiele modulus increases (see Figure 2). This analysis is valid on the assumption that the substrate concentration on the surface of the slab is known and kept constant, as is the case of a continuous stirred tank reactor with the slabs suspended in the solution of substrate. If a fixed bed reactor is to be used, then the substrate concentration is expected to decrease along the reactor as the conversion level increases downstream; in this case, a non-uniform profile of slab thicknesses along the reactor coordinate based on the results plotted in Figure 2 should be considered for maximum net profit if it is feasible to do so. If not, then a weighted average of the values obtained for the optimal slab thickness according to the procedure outlined previously should be used instead.

The shape of the Th_{opt} versus Ma plot shows a steep bend in a narrow range of values for parameter Ma, located at lower levels of Ma as C_s^* gets larger. In the remainder of the scale, the behaviour approaches either a vertical or a horizontal linear asymptote, for lower and higher values of Ma, respectively. In general, the optimum Thiele moduli do not change appreciably with Ma at high values for Ma, this finding being especially true under the condition of high dimensionless surface concentrations. This is equivalent to saying that the actual value of Ma is not too important for the selection of the optimum slab thickness in the diffusional regime. When the kinetic regime (low Th) is reached, however, the optimal slab thickness becomes extremely sensitive to the economic parameter. The analysis reported here can then be ascribed a high value provided that its results are used as a designing tool for the industrial biochemical practice.

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