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Reactor design for the enzymatic isomerization of glucose to fructose

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Abstract. A comprehensive methodology is presented for the design of reactors using immobilized enzymes as catalysts. The design is based on material balances and rate equations for enzyme action and decay and considers the effect of mass transfer limitations on the expression of enzyme activity. The enzymatic isomerization of glucose into fructose with a commercial immobilized glucose isomerase was selected as a case study. Results obtained are consistent with data obtained from existing high-fructose syrup plants. The methodology may be extended to other cases, provided sound expressions for enzyme action and decay are available and a simple flow pattern within the reactor might be assumed.

List of symbols

C	kat/kg	specific activity of the catalyst
D	m^2/s	substrate diffusivity within the catalyst particle
Dr	m	reactor diameter
d	d	operating time of each reactor
E	kat	initial enzyme activity
E_i	kat	initial enzyme activity in each reactor
F	m^3/s	process flowrate
F_i	m^3/s	reactor feed flowrate at a given time
F_0	m^3/s	initial feed flowrate to each reactor
H	–	number of enzyme half-lives used in the reactors
K	$mole/m^3$	equilibrium constant
K_S	$mole/m^3$	Michaelis constant for substrate
K_P	$mole/m^3$	Michaelis constant for product
K_m	$mole/m^3$	apparent Michaelis constant $f(K, K_s, K_p, s_o)$
k	$mole/s \cdot kat$	reaction rate constant
k_d	d^{-1}	first-order thermal inactivation rate constant
L	m	reactor height
L_r	m	height of catalyst bed
N_R	–	number of reactors
P_i	kg	catalyst weight in each reactor
p	$mole/m^3$	product concentration
R	m	particle radius
R_P	–	ratio of minimum to maximum process flowrate
r	m	distance to the center of the spherical particle
s	$mole/m^3$	substrate concentration
s_{0i}	$mole/m^3$	substrate concentration at reactor inlet
s_0	$mole/m^3$	bulk substrate concentration
s'	$mole/m^3$	apparent substrate concentration
T	K	temperature
t	d	time
t_i	d	operating time for reactor i
t_s	d	time elapsed between two successive charges of each reactor
V	m^3	reactor volumen
V_m	$mole/m^3\ s$	maximum apparent reaction rate
V_p	$mole/m^3\ s$	maximum reaction rate for product
V_R	m^3	actual volume of catalyst bed
V_r	m^3	calculated volume of catalyst bed
V_S	$mol/m^3\ s$	maximum reaction rate for substrate
v	$mol/m^3\ s$	initial reaction rate
v_i	m/s	linear velocity
v_m	$mol/m^3\ s$	apparent initial reaction rate $f(K_m, s', V_m)$
X	–	substrate conversion
X_{eq}	–	substrate conversion at equilibrium
$\beta = s/K$	–	dimensionless substrate concentration
$\beta_0 = s_0/K$	–	bulk dimensionless substrate concentration
$\beta_{eq} = s_{eq}/K$	–	dimensionless substrate concentration at equilibrium
η	–	local effectiveness factor
η'	–	mean integrated effectiveness factor
$\theta = \frac{R}{3}\left(\frac{V_m}{K_m D}\right)^{1/2}$	–	Thiéle modulus
$\varrho = r/R$	–	dimensionless radius
ϱ_s	kg/m^3	hydrated support density
σ	–	substrate protection factor
τ	s	residence time

1 Introduction

Fundamentals of enzyme reactor design have been treated by several authors, pointing out different aspects considered relevant [1–4]. However, in very few cases a systematic design procedure has been applied to a particular situation considering all the major variables involved.

A design scheme for enzyme reactors was developed, aiming to consider most relevant parameters comprehensively.

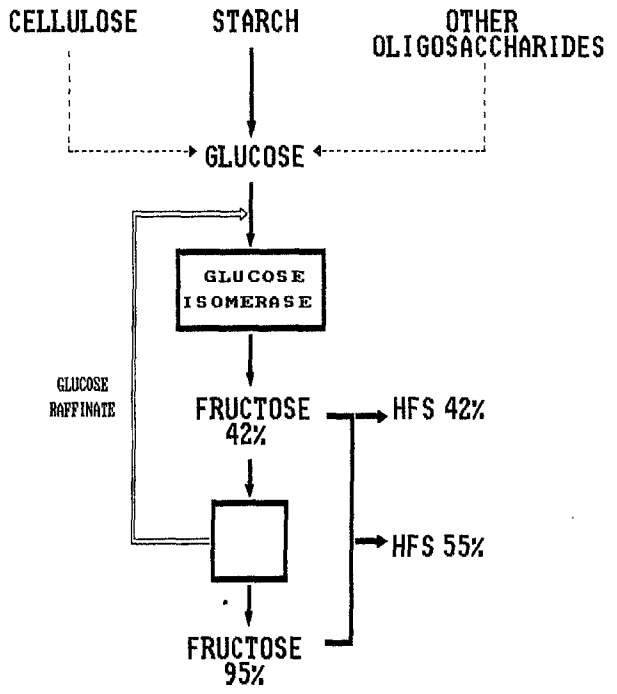

Fig. 1. General scheme for the production of high-fructose syrup from different carbohydrate raw materials

Glucose isomerization with immobilized glucose isomerase was taken as a case study within the context of a plant design for the production of high fructose syrup (HFS) in Chile [5]. HFS production is the most successful industrial application of immobilized enzyme technology. Three main products are currently being produced: 42, 55 and 95% fructose syrups, the former being the product obtained at the enzyme reactor outlet [6, 7]. A general scheme of HFS production starting from different carbohydrate raw materials, is shown in Fig. 1. The plant was designed to produce 18 500 tons/year of 42% HFS (71% solids) and 63 500 tons/year of 55% HFS (77% solids).

2 Design fundamentals

Relevant information for reactor design are the type of enzyme and substrate to be used, operating conditions (temperature, pH, substrate concentration), kinetic model for enzyme reaction and enzyme decay rate, incidence of diffusional restrictions and mode of reactor operation.

Type of reactor can be primarily chosen by kinetic and physicochemical considerations. For continuous operation with immobilized enzymes, column-type reactors operating as fixed, expanded or fluidized bed are often preferred over stirred tank reactors. Packed-bed reactors are mostly used at the industrial scale, because of lesser damage to the enzyme catalyst and ease of retention within the reactor. Temperature profiles and uneven packing of the catalyst can adversely affect packed-bed reactor operation and have to be considered for practical purposes. The kinetics of the enzyme reaction also influences the choice of reactor configuration, packed-bed reactor predicting better results than stirred tank for a reversible Michaelis-Menten type kinetics.

Enzyme catalyst is selected in terms of operational stability and cost per unit of activity expressed.

For the present case, substrate is very much defined. Glucose syrup, 94% dextrose equivalent (DE) and 45% solids will be considered, which is the typical product of the enzymatic liquefaction and saccharification of starch [8].

Operating conditions depend on the enzyme catalyst selected. Reaction temperature is a compromise between enzyme activity and stability. For immobilized glucose isomerase (IGI), temperature optimum lies between 60 to 70 °C. Although we have determined a temperature optimum below 60 °C for the selected catalyst [5], the enzyme supplier suggests not to work below that temperature because of contamination problems during prolonged operation. Therefore 60 °C was selected as the temperature for reactor operation. The pH selected was 7.5, as declared optimum by the supplier [9].

Material balance determines total reactor throughput. Feed flowrate to a particular reactor is determined, for a given amount of enzyme catalyst, by the degree of isomerization required. However, an upper limit exists that prevents backmixing and channeling. The number of reactors is determined to ensure constant degree of isomerization and total throughput within a preestablished margin of variation. The total amount of catalyst required will determine reactor volume. Glucose isomerization to fructose is an equilibrium reaction with an equilibrium constant close to one. Practical substrate conversion for a packed-bed reactor is on the order of 90% of theoretical maximum; therefore a degree of glucose isomerization of 45% was used.

A sound kinetic model is inherent to reactor design. The kinetics of glucose isomerization with IGI is adequately represented by a simple reversible Michaelis-Menten type model. Enzyme deactivation rate at operating conditions needs also to be modelled to predict reactor behaviour. Model should consider substrate protection, to precisely determine catalyst half-life and make-up policy during reactor operation. IGI supplier suggests reactor operation for two catalyst half-lives [9].

Immobilized enzymes are subjected to diffusional restrictions, and its magnitude will depend on the characteristics of the enzyme support, but also on bulk substrate concentration and flow regime. External diffusional restrictions depend on flowrate and could be negligible at values higher than 3 m/s [10]. Internal diffusional restrictions, however, are difficult to overcome for porous matrices, as is the case. This effect can be modelled through the effectiveness factor which depends on substrate concentration, particle size and geometry and inherent kinetic parameters.

3 Operation strategy

Constant reactor throughput of constant product quality is highly desirable for industrial production. Therefore, the

strategy for IGI reaction operation is designed to produce a syrup of constant quality (fructose concentration) with minimum variations in total reactor throughput. Operating at constant temperature, it is possible to obtain constant substrate conversion at the reactor outstream by decreasing flowrate to follow enzyme deactivation rate. Individual reactor throughput will decrase in time, but total throughput can be maintained reasonably constant if several reactors are operated in parallel, but in a programmed disphased mode.

4 Mathematical model

4.1 Development of model

The mathematical model developed permits to determine enzyme decay rate during reactor operation, which allows to profile reactor flowrate for constant degree of isomerization. It also allows the determination of reactor size, number of reactors and mode of operation required for a given allowable variation in product throughput.

The model includes the rate equation, the expression for enzyme inactivation rate under substrate protection, the incidence of diffusional restriction and the material balance.

The enzymatic isomerization of glucose into fructose can be described by a simple reversible Michaelis-Menten rate expression [2]:

$$v = \frac{\frac{V_S \cdot s}{K_S} - \frac{V_P \cdot p}{K_P}}{1 + \frac{s}{K_S} + \frac{p}{K_S}} \tag{1}$$

Starting from a differential material balance, the behaviour of a packed-bed reactor with an immobilized enzyme, assuming piston-flow regime, can be expressed as [1]:

$$\int \frac{dX}{\eta'(X) \cdot v(X)} = \frac{\tau}{s_{0i}} \tag{2}$$

4.2 Evaluation of diffusional restrictions within the catalyst particle

The effect of diffusional restrictions is represented by the effectiveness factor $\eta(X)$ of the catalyst particle. The effectiveness factor has been evaluated in the literature for different particle geometries for simple Michaelis-Menten type kinetics [12–15]; however, no information was available for reversible type kinetics. Eq. (1) can be modified, however, to the mathematical form of a simple irreversible Michaelis-Menten rate expression:

$$v_m = \frac{V_m \cdot s'}{K_m + s'} \tag{3}$$

where:

$$s' = s_{0i}(X_{eq} - X) \tag{4}$$

$$K_m = \frac{K_S K_P}{K_S - K_P}\left[1 + \left(\frac{1}{K_S} + \frac{1}{K_P}\right) \cdot \frac{s_{0i}}{1+K}\right] \tag{5}$$

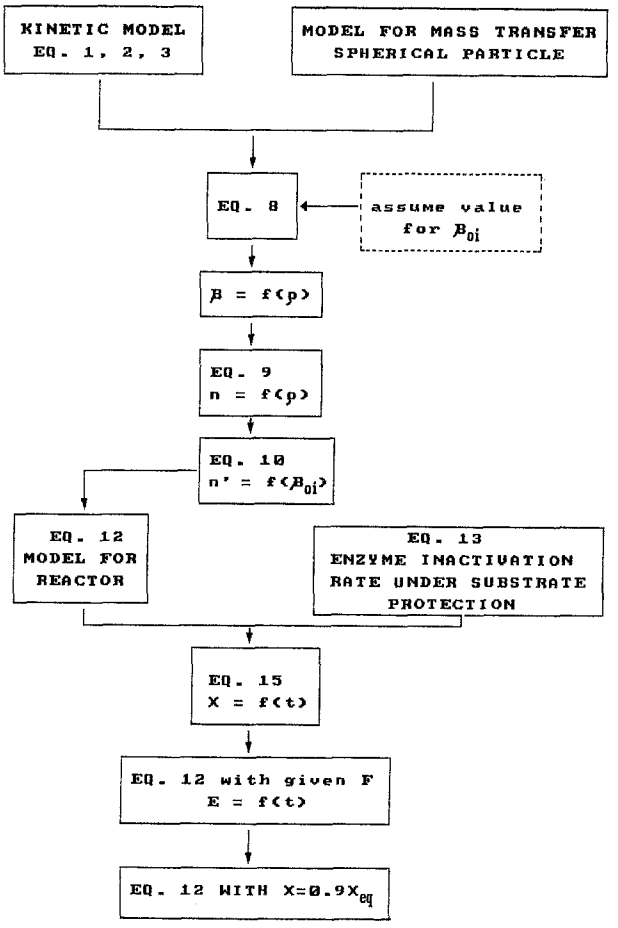

Fig. 2. Scheme for the evaluations of reactor performance and design

$$V_m = \left(1 + \frac{1}{K}\right)\frac{K_p V_s}{K_s - K_p} \tag{6}$$

$$K = \frac{V_S K_P}{V_P K_S} \tag{7}$$

For spherical catalyst particles, a differential analysis, considering Eq. (3) as the rate expression renders:

$$\frac{d^2\beta}{d\varrho^2} + \frac{2}{\varrho}\frac{d\beta}{d\varrho} - 9\theta^2 \frac{\beta - \beta_{eq}}{1 + \beta - \beta_{eq}} = 0 \tag{8}$$

From Eq. (8) the profile of substrate concentration within the catalyst particle is determined; then, the effectiveness factor η is evaluated in each points as:

$$\eta = \frac{(\beta - \beta_{eq})(1 + \beta_0)}{\beta_0(1 + \beta - \beta_{eq})} \tag{9}$$

Then, the mean integrated value of the effectiveness factor η' is evaluated for the catalyst particles as:

$$\eta' = \frac{\int_0^1 \eta \varrho^2 \, d\varrho}{\int_0^1 \varrho^2 \, d\varrho} \tag{10}$$

A computer program evaluates the substrate profile, using fourth-order Runge-Kutta, then evaluates η and then, numerically, the value of η', as shown schematically in Fig. 2.

4.3 Reactor performance considering enzyme decay and diffusional restrictions

The value of η' is dependant on bulk substrate concentration, therefore it varies from point to point within the packed-bed reactor, as shown by the expression $\eta'(X)$ in Eq. (2). As shown in the next section for the case study, $\eta'(X)$ was not a sharp function of X, therefore Eq. (2) could be simplified to:

$$\int \frac{dX}{v(X)} = \eta' \frac{\tau}{s_{0i}} \tag{11}$$

which, for the present case, renders:

$$E = \frac{F K_m}{k \eta'} \left[\frac{s_{0i}}{K_m} X - \ln \left(\frac{X_{eq} - X}{X_{eq}} \right) \right] \tag{12}$$

Enzyme decay rate for IGI under substrate protection has been described in the literature as [16]:

$$\frac{dE}{dt} = (1-\sigma) k_d E \tag{13}$$

where the substrate protection factor σ is:

$$\sigma = \frac{0.5 K_P \left(1 + \frac{s'(1+K)}{s_{0i}}\right) + K_S K \left(1 - \frac{s'(1+K)}{K s_{0i}}\right)}{K_S K_P \frac{(1+K)}{s_{0i}} + K_P \left(1 + \frac{s'(1+K)}{s_{0i}}\right) + K_S K \left(1 - \frac{s'(1+K)}{K s_{0i}}\right)} \tag{14}$$

From Eqs. (12) and (13), Eq. (15) is obtained, which describes reactor operation:

$$\frac{dX}{dt} = \frac{(\sigma - 1) k_d \frac{s_{0i}}{K_m} - \ln \left(\frac{X_{eq} - X}{X_{eq}} \right)}{\frac{s_{0i}}{K_m} + \frac{X_{eq}}{X_{eq} - X}} \tag{15}$$

Equation (15) has to be solved numerically at constant temperature and feed substrate concentration to obtain reactor substrate conversion as a function of operation time.

Enzyme decay rate within the reactor can be obtained in Eq. (12), from the substrate conversion profile obtained by solving Eq. (15). It has to be emphasized that Eq. (12) is only valid at steady-state. However, if the enzyme is reasonably stable, as an immobilized enzyme should be, pseudosteady-state assumption is acceptable [11, 12], and Eq. (12) holds true.

Reactor feed flowrate profile for constant substrate conversion can be obtained directly from the enzyme decay profile since, from Eq. (12), E/F is a constant for constant X.

This procedure is presented schematically in Fig. 2.

4.4 Determination of the number of reactors

The number of reactors required will reflect the policy of operation in terms of the level of catalyst utilization, expressed as the number of half-lives (H), and in terms of the level of total reactor throughput fluctuation allowed (R_P). The number of reactors required can be obtained from [14]:

$$R_P = \exp \left(- \frac{H}{N_R} \ln 2 \right) \tag{16}$$

Each reactor will go into operation at a time interval of t_s days, where:

$$t_s = \frac{d}{N_R} \tag{17}$$

with d the total operating time of each reactor.

Total throughput at a given time corresponds to the sum of the feed flowrates to all reactors at that time:

$$F(t) = \sum_{i=1}^{N_R} F_i \tag{18}$$

Flowrate for each individual reactor at a given time will be determined by the initial flowrate and the time of operation as:

$$F_i = F_0 \exp(-k_d t) \tag{19}$$

Therefore:

$$F(t) = F_0 \exp(-k_d t_s) + \exp(-k_d 2 t_s) + \ldots . + \exp(-k_d (N_R - 1) t_s) + \exp(-k_d N_R t_s) \tag{20}$$

Once N_R has been established, it is possible to determine F_0 from Eqs. (18), (19) and (20).

The amount of catalyst required for each reactor, P_i, can be evaluated as:

$$P_i = \frac{k E_i}{C} \tag{21}$$

and the reaction volume determined as:

$$V_r = \frac{P_i}{\varrho_s} \tag{22}$$

5 Results

The methodology has been evaluated for a commercial IGI from Miles (Takasweet, Miles trade mark for IGI). Relevant data was given by the supplier [9]; other information was gathered from the literature or calculated from the corre-

Table 1. Data for IGI reactor design

		Reference
T	60 °C	[5, 9]
pH	7.5	[9]
X	0.45	[14]
C	7.9 mmol/s kg	[5, 9]
D	2.68×10^{-6} m^2/s	[19]
R	6×10^{-3} m	[19]
s_0	2.805×10^3 mol/m^3	[8]
K_S	7×10^2 mol/m^3	[5, 9,16]
K_P	4.5×10^2 mol/m^3	
V_S	4.87 mmol/s kg	
V_P	3.12 mmol/s kg	
K	1	Eq. (7)
X_{eq}	0.5	[15]
K_m	7.815×10^3 mol/m^3	Eq. (5)
V_m	17.52 mmol/s kg	Eq. (6)
η'	0.8	[5]
k_d	8.06×10^{-3} días^{-1}	[5, 9]
F	5.041×10^{-3} m^3/s	[5]
R_P	0.818	[5]
H	2	[9]

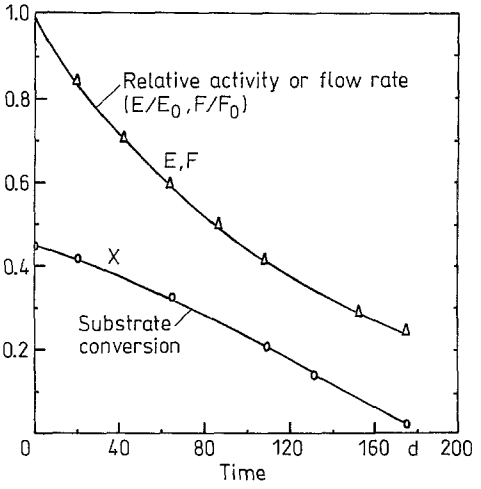

Fig. 3. Results for reactor performance. △: variation of E or F at constant $X = 0.45$; ○: variation of X at constant F

Table 2. Summary of results for IGI reactor design

$t_{1/2}$	86 days	
$d\ (\sim 2\, t_{1/2})$	176 days	
N_R	8	Eq. (16)
t_s	22	Eq. (17)
F_0	4.65 m^3/h	Eq. (19)
P_i	1781 kg	Eq. (21)
V_R	5.32 m^3	Eq. (22)
V	8.19 m^3	
L_r	4.5 m	
L	6.3 m	
D_r	1.286 m	
v_i	3.6 m/h	

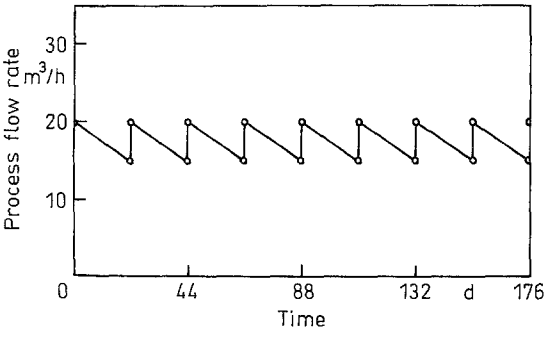

Fig. 4. Results for the operation of the reaction system composed by eight reactors

sponding equations. This information is presented in Table 1.

For reactor design, the following considerations were made, in accordance with the information given by the enzyme supplier:

V_R	$= 1.1\ V_r$	[3, 20]
V	$= 1.4\ V_R$	
L_r/D_r	> 3	[21, 22] (a value of 3.5 was used)
$L_r \times v_i$	< 40 m^2/h	[22]
L_r	< 5 m	[21, 22]

Results showed a mild dependence of η' on bulk substrate concentration, which validated the assumption made in developing Eq. (11). Therefore, η' was considered constant and the average value of 0.8 was used throughout the calculations.

Enzyme decay and substrate conversion during reactor operation at constant feed flowrate were calculated from Eqs. (12) to (15) for the data gathered in Table 1. Results are presented in Fig. 3. Feed flowrate profile for a constant substrate conversion of 0.45 is represented by the same curve of enzyme decay in Fig. 3, as predicted by Eq. (12).

The plant was designed to produce 18 500 tons/year of 42% HFS (71% solids) and 63 500 tons/year of 55% HFS (77% solids). According to this throughput, the total number of reactors, reactor sequencing, catalyst charge and reactor dimensions were calculated according to Eqs. (16) to (22). Results are summarized in Table.2.

Reactor operation is depicted in Fig. 4 for the eight units operating simultaneously but disphased 22 days from each other. Calculated enzyme yield was 0.41 kg/ton of solids treated, which is within the values reported in the literature for similar plants, which range between 0.3 and 0.55 kg/ton [21, 22].

6 Conclusions

The methodology may prove useful for the design or evaluation of reactor performance with immobilized enzymes sub-

jected to diffusional restrictions and activity losses during operation. The analysis may be extended to other kinetic models, enzyme decay rate expressions and reactor configuration, provided a simple flow pattern can be established. Significant $\eta'(X)$ profiles within the reactor may be included in solving Eq. (2), which will only add to mathematical complexity of the calculation scheme. This was considered irrelevant for the present case and remains to be done for those cases in which significant η' profiles exist along the catalyst bed.

Results for the case study of enzymatic isomerization of glucose syrup with IGI are consistent with data from HFS producing plants.

Diffusional restrictions were not significant and the effectiveness factor depended on substrate concentration only mildly in the range of interest. The effect of substrate protection on enzyme decay rate, although seldom considered, proved to be significant, increasing operational enzyme half-life to more than 70%.

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