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Bioprocess Engineering Principles

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II

Homogeneous Reactions

The heart of a typical bioprocess is the reactor or fermenter. Flanked by unit operations which carry out physical changes for medium preparation and recovery of products, the reactor is where the major chemical and biochemical transformations occur. In many bioprocesses, characteristics of the reaction determine to a large extent the economic feasibility of the project.

Of most interest in biological systems are *catalytic* reactions. By definition, a catalyst is a substance which affects the rate of reaction without altering the reaction equilibrium or undergoing permanent change itself. Enzymes, enzyme complexes, cell organelles and whole cells perform catalytic roles; the latter may be viable or non-viable, growing or non-growing. Biocatalysts can be of microbial, plant or animal origin. Cell growth is an *autocatalytic reaction*: this means that the catalyst is a product of the reaction. The performance of catalytic reactions is characterised by variables such as the reaction rate and yield of product from substrate. These parameters must be taken into account when designing and operating reactors.

In engineering analysis of catalytic reactions, a distinction is made between *homogeneous* and *heterogeneous* reactions. A reaction is homogeneous if the temperature and all concentrations in the system are uniform. Most fermentations and enzyme reactions carried out in mixed vessels fall into this category. In contrast, heterogeneous reactions take place in the presence of concentration or temperature gradients. Analysis of heterogeneous reactions requires application of mass-transfer principles in conjunction with reaction theory. Heterogeneous reactions are treated in Chapter 12.

This chapter covers the basic aspects of reaction theory which allow us to quantify the extent and speed of homogeneous reactions and to identify important factors affecting reaction rate.

11.1 Basic Reaction Theory

Reaction theory has two fundamental parts: *reaction thermodynamics* and *reaction kinetics*. Reaction thermodynamics is concerned with *how far* the reaction can proceed; no matter how fast a reaction is, it cannot continue beyond the point of chemical equilibrium. On the other hand, reaction kinetics is concerned with the *rate* at which reactions proceed.

11.1.1 Reaction Thermodynamics

Consider a reversible reaction represented by the following equation:

$$A + bB \rightleftharpoons yY + zZ. \tag{11.1}$$

A, B, Y and Z are chemical species; b, y and z are stoichiometric coefficients. If the components are left in a closed system for an infinite period of time, the reaction proceeds until *thermodynamic equilibrium* is reached. At equilibrium there is no net driving force for further change; the reaction has reached the limit of its capacity for chemical transformation in a closed system. Composition of the equilibrium mixture is determined exclusively by the thermodynamic properties of the reactants and products; it is independent of the way the reaction is executed. Equilibrium concentrations are related by the *equilibrium constant*, K. For the reaction of Eq. (11.1):

$$K = \frac{C_{Ye}{}^{y} C_{Ze}{}^{z}}{C_{Ae}\, C_{Be}{}^{b}} \tag{11.2}$$

where C_{Ae}, C_{Be}, C_{Ye} and C_{Ze} are equilibrium concentrations of A, B, Y and Z, respectively. The value of K depends on temperature as follows:

$$\ln K = \frac{-\Delta G^{\circ}_{rxn}}{RT} \tag{11.3}$$

where ΔG°_{rxn} is the *change in standard free energy* per mole of A reacted, R is the ideal gas constant and T is absolute temperature. Values of R are listed in Table 2.5 (p. 20). The superscript ° in ΔG°_{rxn} indicates standard conditions. Usually,

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the standard condition for a substance is its most stable form at 1 atm pressure and 25°C; however, for biochemical reactions occurring in solution, other standard conditions may be used [1]. ΔG°_{rxn} is equal to the difference in *standard free energy of formation*, G°, between products and reactants:

$$\Delta G^\circ_{rxn} = y\,G^\circ_Y + z\,G^\circ_Z - G^\circ_A - b\,G^\circ_B\,. \tag{11.4}$$

Standard free energies of formation are available in handbooks such as those listed in Section 2.6.

Free energy G is related to enthalpy H, entropy S and absolute temperature T as follows:

$$\Delta G = \Delta H - T\Delta S. \tag{11.5}$$

Therefore, from Eq. (11.3):

$$\ln K = \frac{-\Delta H^\circ_{rxn}}{RT} + \frac{\Delta S^\circ_{rxn}}{R}. \tag{11.6}$$

Thus, for exothermic reactions with negative ΔH°_{rxn}, K decreases with increasing temperature. For endothermic reactions and positive ΔH°_{rxn}, K increases with temperature.

Example 11.1 Effect of temperature on glucose isomerisation

Glucose isomerase is used extensively in the USA for production of high-fructose syrup. The reaction is:

glucose $\rightleftharpoons$ fructose.

ΔH°_{rxn} for this reaction is 5.73 kJ gmol^{-1}; ΔS°_{rxn} is 0.0176 kJ gmol^{-1} K^{-1}.

(a) Calculate the equilibrium constants at 50°C and 75°C.
(b) A company aims to develop a sweeter mixture of sugars, i.e. one with a higher concentration of fructose. Considering equilibrium only, would it be more desirable to operate the reaction at 50°C or 75°C?

Solution:
(a) Convert temperatures to degrees Kelvin (K) using the formula of Eq. (2.24):

$T = 50°C = 323.15$ K
$T = 75°C = 348.15$ K.

From Table 2.5, $R = 8.3144$ J gmol^{-1} K^{-1} $= 8.3144 \times 10^{-3}$ kJ gmol^{-1} K^{-1}. Using Eq. (11.6)

$$\ln K\,(50°C) = \frac{-5.73\ \text{kJ gmol}^{-1}}{(8.3144 \times 10^{-3}\ \text{kJ gmol}^{-1}\,\text{K}^{-1})\,(323.15\text{K})} + \frac{0.0176\ \text{kJ gmol}^{-1}\,\text{K}^{-1}}{8.3144 \times 10^{-3}\ \text{kJ gmol}^{-1}\,\text{K}^{-1}}$$

$$K\,(50°C) = 0.98.$$

Similarly for $T = 75°C$:

$$\ln K\,(75°C) = \frac{-5.73\ \text{kJ gmol}^{-1}}{(8.3144 \times 10^{-3}\ \text{kJ gmol}^{-1}\,\text{K}^{-1})\,(348.15\text{K})} + \frac{0.0176\ \text{kJ gmol}^{-1}\,\text{K}^{-1}}{8.3144 \times 10^{-3}\ \text{kJ gmol}^{-1}\,\text{K}^{-1}}$$

$$K\,(75°C) = 1.15.$$

(b) As K increases, the fraction of fructose in the equilibrium mixture increases. Therefore, from an equilibrium point of view, it is more desirable to operate the reactor at 75°C. However, other factors such as enzyme deactivation at high temperatures should also be considered.

A limited number of commercially-important enzyme conversions, such as glucose isomerisation and starch hydrolysis, are treated as reversible reactions. In these systems, the reaction mixture at equilibrium contains significant amounts of reactants as well as products. However, for many reactions ΔG°_{rxn} is negative and large in magnitude. As a result, K is also very large, the reaction favours the products rather than the reactants, and the reaction is regarded as *irreversible*. Most enzyme and cell reactions fall into this category. For example, the equilibrium constant for sucrose hydrolysis by invertase is about 10^4; for fermentation of glucose to ethanol and carbon dioxide, K is about 10^{30}. The equilibrium ratio of products to reactants is so overwhelmingly large for these reactions that they are considered to proceed to completion, i.e. the reaction stops only when the concentration of one of the reactants falls to zero. Equilibrium thermodynamics has therefore only limited application to enzyme and cell reactions. Moreover, the thermodynamic principles outlined in this section apply only to closed systems; true thermodynamic equilibrium does not exist in living cells which exchange matter with their surroundings. Metabolic processes in cells are in a dynamic state; products formed are constantly removed or broken down so that reactions are driven forward. Most reactions in biological systems proceed to completion in a finite period of time at a finite rate.

If we know that complete conversion will eventually take place, the most useful reaction parameter to know is the rate at which the transformation proceeds. Another important characteristic, especially for systems in which many different reactions take place at the same time, is the proportion of reactant that is converted to the desired products. These properties of reactions are discussed in the remainder of this chapter.

11.1.2 Reaction Yield

The extent to which reactants are converted to products is expressed as the reaction *yield*. Generally speaking, yield is the amount of product formed or accumulated per amount of reactant provided or consumed. Unfortunately, there is no strict definition of yield; several different yield parameters are applicable in different situations. The terms used to express yield in this text do not necessarily have universal acceptance and are defined here for our convenience. Be prepared for other books to use different definitions.

Consider the simple enzyme reaction:

$$\text{L-histidine} \rightarrow \text{urocanic acid} + NH_3 \tag{11.7}$$

catalysed by histidase. According to the reaction stoichiometry, 1 gmol urocanic acid is produced for each gmol L-histidine consumed; the yield of urocanic acid from histidine is therefore 1 gmol gmol^{-1}. However, let us assume that the histidase used in this reaction is contaminated with another enzyme, histidine decarboxylase. Histidine decarboxylase catalyses the following reaction:

$$\text{L-histidine} \rightarrow \text{histamine} + CO_2. \tag{11.8}$$

If both enzymes are active, some L-histidine will react with histidase according to Eq. (11.7), while some will be decarboxylated according to Eq. (11.8). After addition of the enzymes to the substrate, analysis of the reaction mixture shows that 1 gmol urocanic acid and 1 gmol histamine are produced for every 2 gmol histidine consumed. The *observed* or *apparent yield* of urocanic acid from L-histidine is $^{1\ \text{gmol}}/_{2\ \text{gmol}} = 0.5$ gmol gmol^{-1}. The observed yield of 0.5 gmol gmol^{-1} is different from the *stoichiometric*, *true* or *theoretical yield* of 1 gmol gmol^{-1} calculated from reaction stoichiometry because the reactant was channelled in two separate reaction pathways. An analogous situation arises if product rather that reactant is consumed in other reactions; the observed yield of product would be lower than the theoretical yield. *When reactants or products are involved in additional reactions, the observed yield may be different from the theoretical yield.*

The above analysis leads to two useful definitions of yield for reaction systems:

$$\begin{pmatrix}\text{true, stoichiometric or}\\ \text{theoretical yield}\end{pmatrix} = \frac{\begin{pmatrix}\text{total mass or moles of}\\ \text{product formed}\end{pmatrix}}{\begin{pmatrix}\text{mass or moles of reactant used}\\ \text{to form that particular product}\end{pmatrix}} \tag{11.9}$$

and

$$\begin{pmatrix}\text{observed or}\\ \text{apparent yield}\end{pmatrix} = \frac{(\text{mass or moles of product present})}{\begin{pmatrix}\text{total mass or moles of reactant}\\ \text{consumed}\end{pmatrix}}. \tag{11.10}$$

There is a third type of yield applicable in certain situations. For reactions with incomplete conversion of reactant, it may be of interest to specify the amount of product formed per amount of reactant *provided to the reaction* rather than actually consumed. For example, consider the isomerisation reaction catalysed by glucose isomerase:

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