Diffusional Limitations in Immobilized Enzyme System

- Immobilized enzyme system normally includes
  - insoluble immobilized enzyme
  - soluble substrate, or product
- They are heterogeneous systems

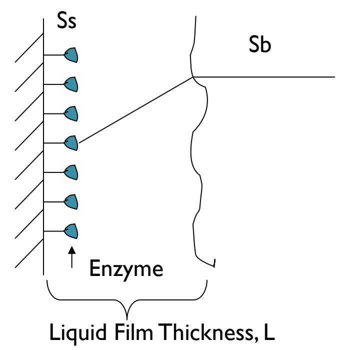
Diffusional Limitation in Immobilized Enzyme Systems

- In immobilized enzyme systems,
  the overall production rate is determined by
  - **liquid film mass transfer** (external diffusion) substrate, product
  - **intraparticle mass transfer** (internal diffusion) substrate, product in porous supports
  - **enzyme catalysis reaction**

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

$$E + S \rightleftarrows ES \xrightarrow{k_2} P + E$$

Assume the enzyme catalyzed reaction rate follows Michaelis-Menten type kinetics.



Ss: substrate concentration at the surface;
Sb: substrate concentration in bulk solution.

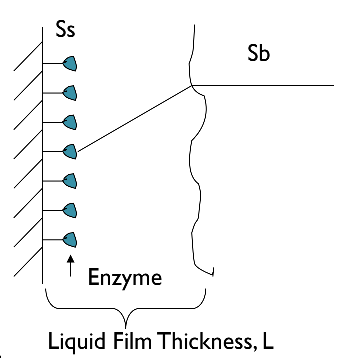
Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

Assume:

- Enzyme are evenly distributed on the surface of a nonporous support material.

- All enzyme molecules are equally active.

- Substrate diffuses through a thin liquid film surrounding the support surface to reach the reactive surface.



No intraparticle diffusion

-The process of immobilization has not altered the enzyme structure and the intrinsic parameters (V_m, K_m).

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

To determine the significant effect of external diffusion resistance on the rate of enzyme catalytic reaction rate: **Damköhler numbers** (Da)

$$Da = \frac{\text{maximum rate of reaction}}{\text{maximum rate of diffusion}} = \frac{V_m'}{k_L[S_b]}$$

V_m' = the maximum reaction rate per unit of external surface area (e.g. g/cm^2-s)

k_L = the liquid mass transfer coefficient (cm/s)

$[S_b]$ = the substrate concentration in bulk solution (g/cm^3)

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

$$Da = \frac{\text{maximum rate of reaction}}{\text{maximum rate of diffusion}} = \frac{V_m'}{k_L[S_b]}$$

When

- Da >> 1, the external diffusion rate is limiting;
- Da << 1, the reaction rate is limiting;
- Da ≈ 1, the external diffusion and reaction resistances are comparable.

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

The external diffusion rate J_S (g/cm^2-s):

$$J_s = k_L([S_b]-[S_s])$$

k_L is the liquid mass transfer coefficient (cm/s).

If the product formation rate is :

$$v' = \frac{V_m'[S_s]}{K_m+[S_s]}$$

V_m' is the maximum reaction rate per unit surface area (g/cm^2-s).

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

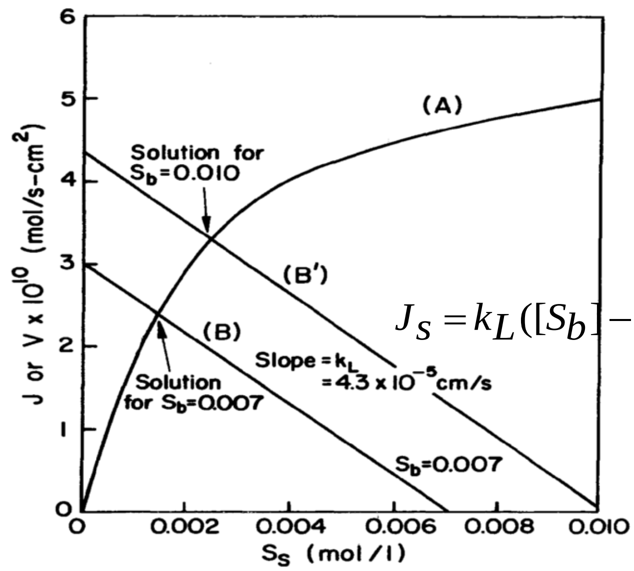
$$E + S \rightleftarrows ES \xrightarrow{k_2} P + E$$

At steady state, the reaction rate is equal to the external diffusion rate:

$$J_s = k_L([S_b]-[S_s]) = \frac{V_m'[S_s]}{K_m+[S_s]}$$

With the equation and known S_b, K_L, V_m' or K_m, to determine numerically or graphically:

- The substrate concentration at the surface.
- The reaction rate.



$$J_S = k_L([S_b] - [S_S])$$

Graphical solution for reaction rate per unit of surface area for enzyme immobilized on a non-porous support

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

When the system is strongly external diffusion (liquid film mass-transfer) limited, [Ss]≈0, the overall reaction rate is equal to the rate:

$$v = k_L[S_b] \qquad Da >> 1$$

The system behaves as pseudo first order.

The rate is a linear function of bulk substrate concentration.

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

To increase the overall reaction rate with external diffusion limitation

$$Da = \frac{\text{maximum rate of reaction}}{\text{maximum rate of diffusion}} = \frac{V_m'}{k_L[S_b]}$$

-Increase ________________________________.

-Increase ________________________________.

The liquid film mass transfer coefficient k_L:

$$k_L \approx 0.6 \times \left(\frac{D_{AB}^{2/3}}{\nu^{1/6}} \right) \times \left(\frac{U^{1/2}}{d_p^{1/2}} \right)$$

(H. Fogler, Elements of Chemical Reaction Engineering 1999, p705)

D_{AB} = mass diffusivity of the substrate in the liquid phase, a function of temperature and pressure (m^2/s)
ν = kinematic viscosity (m^2/s), a function of temperature.
U = free-system liquid velocity
(velocity of the fluid flowing past the particle) (m/s).
d_p = size of immobilized enzyme particle (m).

At specific T and P, increasing U and decreasing d_p increase the liquid film mass transfer coefficient and thus the external diffusion rate.

Diffusion Effects in Surface-bound Enzymes on Nonporous Support Materials

When the system is strongly reaction limited,

$$[S_b] \approx [S_s]$$

and the overall reaction rate is equal to the rate:

$$v = \frac{V_m'[S_b]}{K_{m,app} + [S_b]} \qquad Da << 1$$

where

$$K_{m,app} = K_m \left\{ 1 + \frac{V_m'}{k_L([S_b] + K_m)} \right\}$$

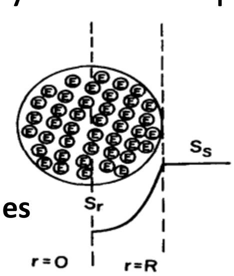
$K_{m,app}$ is increased.
It is a function of mixing speed and S_b.

Diffusion Effects in Enzymes Immobilized in a Porous Matrix

- Substrate diffuses through the tortuous pathway within the porous support to reach the enzyme.

- Substrate reacts with enzyme on the pore surface.

Ex. Spherical support particles



Diffusion Effects in Enzymes Immobilized in a Porous Matrix

- Assume:
  - Enzyme is uniformly distributed in a spherical support particle.
  - The reaction kinetics follows Michaelis-Menten kinetics.
  - There is no external diffusion limitation.

Under internal diffusion limitations, the rate per unit volume is expressed in terms of the effectiveness factor as follows:

$$r_s = \eta \frac{V_m''[S_s]}{K_m + [S_s]}$$

η = effectiveness factor.

V_m'' = maximum velocity per volume of the support.

K_m = M-M constant.

$[S_s]$ = substrate concentration on the surface of the support.

$$\eta = \frac{\text{reaction rate with intraparticle diffusion limitation}}{\text{reaction rate without diffusion limitation}}$$

$\eta < 1$ the rate is diffusion limited.

$\eta = 1$ the rate is reaction rate limited.

Diffusion Effects in Enzymes Immobilized in a Porous Matrix

$$\eta = \frac{3}{\phi}\left[\frac{1}{\tanh\phi} - \frac{1}{\phi}\right]$$

$$\phi = R\sqrt{\frac{V_m''/K_m}{D_e}}$$ =Thiele modulus

D_e = effective diffusivity of substrate within the porous matrix

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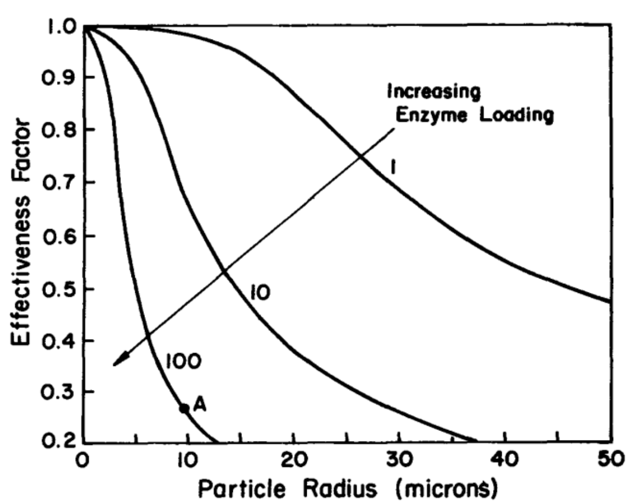

Relationship of effectiveness factor with the size of immobilized enzyme particle and enzyme loading

Diffusion Effects in Enzymes Immobilized in a Porous Matrix

- At specific conditions (T, P) for a fixed system, to increase the intra-particle mass transfer rate:

  - ________ the size of immobilized enzyme particle

  - _________ the porosity or specific surface area of the particle