BIOLOGICAL REACTIONS AND BIOREACTORS: FERMENTATIONS

Biological reactions are more complex than anything that we have considered so far and the reasons for this are briefly outlined below. Consider a “generic” biological reaction where some organic matter (food) is consumed by a culture of living organisms (bugs) that also consume oxygen due to their respiration (aerobic culture) and in the process generate carbon dioxide, water and more biomass (more bugs). This is represented by equation (1):

$$\text{Organic matter} + O_2 + \text{bugs} \rightarrow CO_2 + H_2O + \text{more bugs} \quad (1)$$

This process involves oxidation, respiration and synthesis (of new biomass).

Oxidation is the electron donor half reaction, shown by equation (2), with rate $R_d$.

$$\frac{1}{24} C_6H_{12}O_6 + \frac{1}{4} H_2O \rightarrow \frac{1}{4} CO_2 + H^+ + e^-; \quad R_d \quad (2)$$

Reduction is the electron acceptor half reaction that has two possible routes: respiration, equation (3a), and synthesis, equation (3b), that proceed at the rate $R_c$ and $R_s$, respectively.

A. Respiration

$$\frac{1}{4} O_2 + H^+ + e^- \rightarrow \frac{1}{2} H_2O; \quad R_c \quad (3a)$$

B. Synthesis (in presence of nitrates)

$$\frac{1}{28} NO_3^- + \frac{5}{22} CO_2 + \frac{19}{28} H^+ + e^- \rightarrow \frac{1}{28} C_5H_7NO_2 + \frac{11}{28} H_2O; \quad R_s \quad (3b)$$

In the above we have assumed that organic matter (food) is a simple sugar (glucose) and that the culture in question can utilize the nitrate ion to synthesize new biomass by equation (3b). Now, we assume that the probability of reaction (3a) occurring is $f_c$ while the probability of reaction (3b) occurring is $f_s$. Clearly, since the electron must be used by either route (3a) or (3b), we must have $f_c + f_s = 1$. We need data to determine $f_s$ or $f_c$ or preferably both! In addition, the rate of electron production must equal the rate of electron consumption so that

$$R_d + f_s R_s + f_c R_c = 0 \quad (4)$$

Now, if based on available information we conclude that $f_s = 0.62$, and hence $f_c = 1 - 0.62 = 0.38$, then substituting these values in equation (4) leads to the overall stoichiometry below.
Overall stoichiometry

\[
\frac{1}{24} C_6H_{12}O_6 + 0.0221NO_3^- + 0.095O_2 + 0.0221H^+ \rightarrow 0.0221C_5H_7NO_2 + 0.14CO_2 + 0.184H_2O
\]  
(5)

Now, if we know the O\(_2\) uptake rate we can determine the nitrate and/or glucose consumption rate and rate of synthesis of cell mass! Unfortunately, in most real complex systems we do not know that and we need to rely on empirical yield factors which will be defined below.

We need now to describe the rate of biomass (bugs) growth, substrate (food, e.g. sugar) consumption rate and oxygen consumption rate.

**Biomass Growth Rate**

Typically, Monod’s model is used where the rate of formation of the biomass, \(R_x\), is proportional to the biomass concentration present in the system, \(X\), as shown by equation (6)

\[
R_x = \mu X
\]  
(6)

The specific growth constant is given by Michaelis Menten kinetic form and depends on the limiting substrate concentration, \(S\), as shown below.

\[
\mu = \frac{\mu_m S}{K_s + S}
\]  
(7)

The above ignores the death term in the cell rate which has to be included for mature cultures, so that a more complete description of the net biomass formation rate is:

\[
R_s = \frac{\mu_m S X}{K_s + S} - k_d X
\]  
(8)

**Substrate Consumption Rate**

The substrate consumption rate is customarily given in terms of the biomass production rate and the appropriate yield coefficient. Please, note that the yield coefficient may change with conditions but is most frequently used as constant:

\[
-R_s = \frac{1}{Y_{x/s}} R_x
\]  
(9)
\[ Y_{x/s} = \text{yield coefficient} = \frac{(mg/L) \text{biomass produced}}{(mg/L) \text{substrate consumed}} \]  
(10)

**Oxygen Consumption Rate**

This rate must also be expressed in terms of an empirical yield coefficient as shown below:

\[-R_o = \frac{1}{Y_{x/o}} R_x \]  
(11)

\[ Y_{x/o} = \frac{(mg/L) \text{biomass produced}}{(mg/L) \text{oxygen consumed}} \]  
(12)

**Balance on well mixed compartment (CSTR at transient conditions)**

Well mixed CSTR at unsteady state can now be described by writing the mass balance on biomass, substrate and oxygen as follows:

\[ V \frac{dX}{dt} = Q(X_o - X) + R_x V \]  
(13)

\[ t = 0 \quad X = X_i \]

\[ V \frac{dS}{dt} = Q(S_o - S) - (-R_s)V \]  
(14)

\[ t = 0 \quad S = S_i \]

\[ V \frac{dC_o}{dt} = Q(C_o^* - C_o) + k_s a V(C_o^* - C_o) - (-R_o)V \]  
(15)

\[ t = 0 \quad C_o = C_{oi} \]

Dilution rate (space velocity in CHE jargon) is denoted by:

\[ D = \frac{Q}{V} \]  
(16)

Then, rewriting equation (13) to (15) above yields:
\[
\frac{dX}{dt} = D(X_0 - X) + \frac{\mu_m S}{K_s + S} X 
\]  \hspace{1cm} (17)

\[
\frac{dS}{dt} = D(S_0 - S) - \frac{1}{Y_{s/l}} \frac{\mu_m S}{K_s + S} X 
\]  \hspace{1cm} (18)

\[
\frac{dC_o}{dt} = D(C_0 - C_o) - \frac{1}{Y_{s/l,o}} \frac{\mu_m S}{K_s + S} X + k_i \alpha(C_0^* - C_o) 
\]  \hspace{1cm} (19)

In a batch system \(D = 0\).

In a steady state CSTR \(\frac{d}{dt} = 0\). Let us look at a steady state CSTR.

A steady state CSTR has the advantage that can operate with no fresh culture in the feed stream, \(X_0 = 0\), so that from equation (17) see that the dilution rate is balanced by the specific growth rate as given by equation (20)

\[ D = \mu \]  \hspace{1cm} (20)

\[ D = \frac{\mu_m S}{K_s + S} \rightarrow \text{Solve for } S \]  \hspace{1cm} (21)

\[ S = \frac{K_s D}{\mu_m - D} \quad \text{for } D < D_{cr} \]  \hspace{1cm} (22)

In case that the dilution rate is larger than the critical dilution rate given below, washout occurs and only the steady state at zero biomass concentration is possible.

\[ D > D_{cr} = \frac{\mu_m S_o}{K_s + S_o} \Rightarrow X = 0 \]  \hspace{1cm} (23)

For admissible values of the dilution rate \(D\) the biomass steady state concentration in the effluent (and hence in the reactor due to CSTR assumptions) is given by

\[ X = X_{ss} = Y_{s/l} (S_0 - S) = Y_{s/l} \left[ S_0 - \frac{K_s D}{\mu_m - D} \right] \]  \hspace{1cm} (24)

The production rate of the biomass per unit reactor volume is the product \(DX\)

To maximize it we need
\[
\frac{d}{dD} (DX) = 0 \text{ which yields:}
\]
\[
D_{opt} = \mu_m \left[ 1 - \left( \frac{K_s}{K_s + S_o} \right)^{1/2} \right]
\]
(25)

And produces the following cell concentration in the effluent
\[
X_{opt} = \frac{Y_{x/s}}{S_o + K_s - \left( \frac{K_s}{S_o + K_s} \right)^{1/2}}
\]
(26)

See diagram below: