Stoichiometric coefficients for growth

Yield coefficients, Y, are defined based on the amount of consumption of another material

Growth Yield $$Y_{X/S} = -\frac{\Delta X}{\Delta S}$$

$$\Delta S = \Delta S_{\text{assimilated into biomass}} + \Delta S_{\text{assimilated into extracellular products}} + \Delta S_{\text{growth energy}} + \Delta S_{\text{maintenance energy}}$$

Because ΔS changes with growth condition, $Y_{X/S}$ is not a constant

Stoichiometric coefficients for growth (cont.)

Typical range of yield coefficients

Growth Yield $$Y_{X/S} = -\frac{\Delta X}{\Delta S}$$

$Y_{X/S} \approx$ 0.4 to 0.6 g dry cells/g substrate consumed

$Y_{X/S,\ oxidized\ S}$ (0.4 to 0.6) < $Y_{X/S,\ reduced\ S}$ (0.6 to 1.0)

Other Yield Coefficients: $$Y_{X/O_2} = -\frac{\Delta X}{\Delta O_2}$$

$Y_{X/O_2,\ reduced\ S}$ (0.15 to 0.3) < $Y_{X/O_2,\ oxidized\ S}$ (0.3 to 1.5)

Stoichiometric coefficients for growth (cont.)

Other Yield Coefficients: $Y_{X/ATP} = \frac{\Delta X}{\Delta ATP} \left(\frac{\text{g dry cells}}{\text{mole ATP generated}}\right)$

$Y_{X/ATP}$ > complex medium (a.a.s and nucleic acids available)

$Y_{X/ATP}$ < minimal medium (only inorganic salts and substrate)

Anaerobic Fermentations:

$Y_{X/ATP} \approx 10.5 \pm 2$ (g dry cell wt./mole ATP)

$C_6H_6O_6 \rightarrow 2\ C_2H_5OH + 2\ CO_2$: 2 moles ATP/mole glucose

$Y_{X/S} \approx Y_{X/ATP}\,(2)/MW_{glu} = (10.5)(2)/180 = 0.12$ g dcw/g glucose

Specific production rate (q_p)
Product yield coefficients ($Y_{p/x}$)

1. Growth associated products: products appear simultaneously with cells in culture

$$q_P = \frac{1}{X}\frac{dP}{dt} = Y_{P/X}\,\mu$$

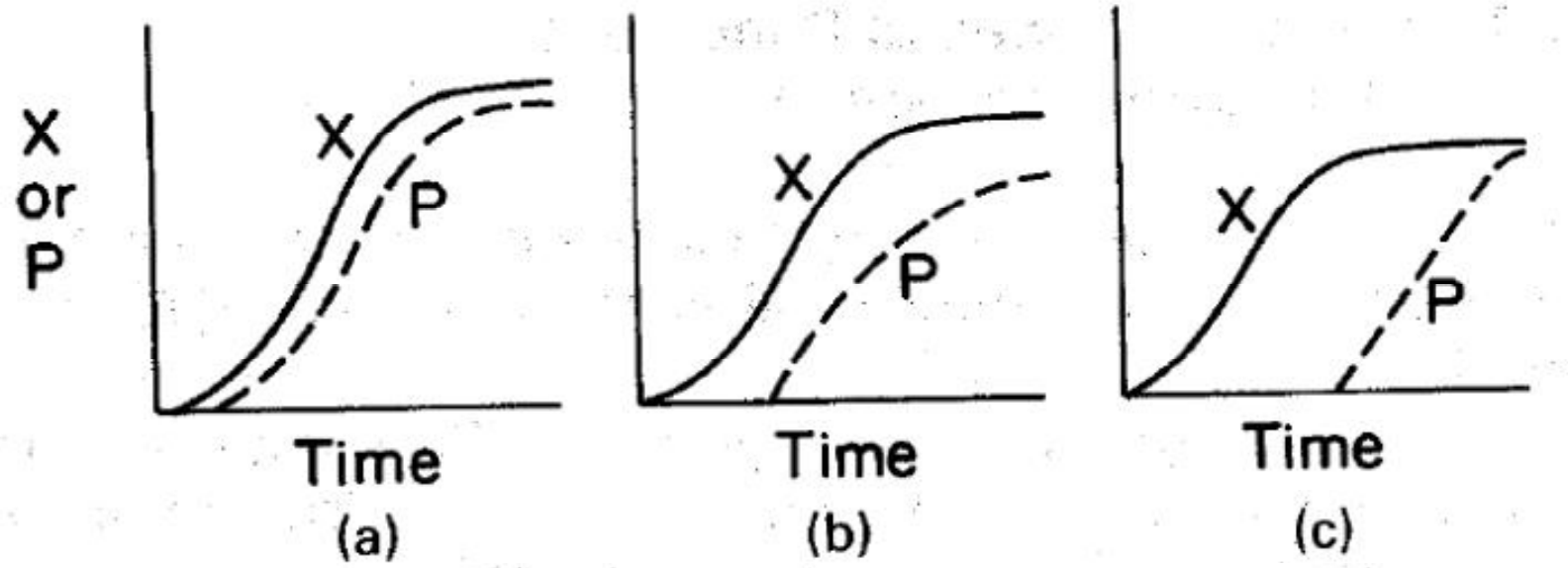
2. Non-growth associated products: products appear during stationary phase of batch growth

$$q_P = \beta$$

3. Mixed-growth associated products: products appear during slow growth and stationary phase

$$q_P = \alpha\mu + \beta$$

Specific production rate (q_p)



$$1.\ q_P = \frac{1}{X}\frac{dP}{dt} = Y_{P/X}\,\mu \qquad 3.\ q_P = \alpha\mu + \beta \qquad 2.\ q_P = \beta$$

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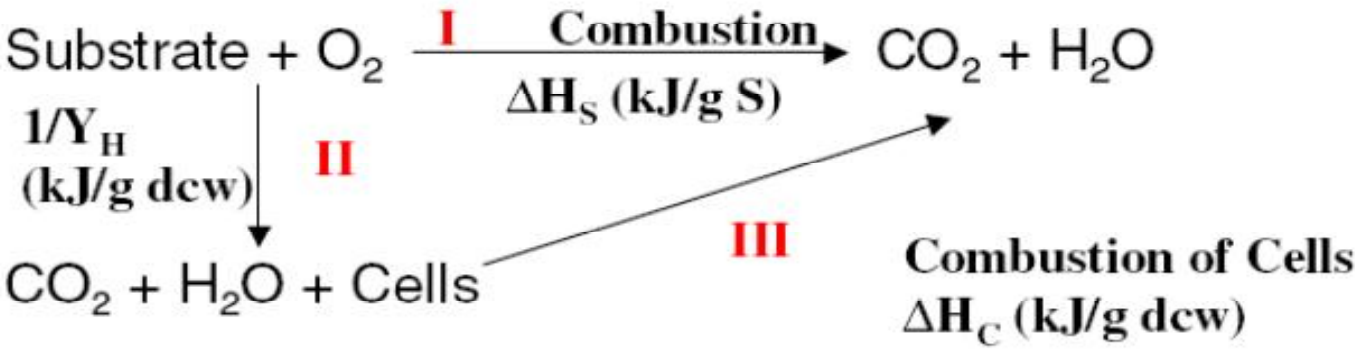
Heat generation by growth

Only 40 to 50% of the energy stored in a carbon substrate is converted to biological energy (ATP) during aerobic metabolism. The remainder is released as heat upon conversion to CO_2 and H_2O

Energy Balance: $\left[\begin{matrix}\text{Total Available}\\ \text{Energy of Substrate}\end{matrix}\right]^{I} = \left[\begin{matrix}\text{Energy Released}\\ \text{by Growth}\end{matrix}\right]^{II} + \left[\begin{matrix}\text{Energy Available}\\ \text{in Biomass}\end{matrix}\right]^{III}$



Heat generation by growth (cont.)

On a per gram of substrate basis

$$(1\ g\ S)\ \Delta H_S = (1\ g\ S)\ Y_{X/S}/Y_H + (1\ g\ S)\ Y_{X/S}\ \Delta H_C$$

Heat generated during cell growth

Energy in biomass

Solving for Y_H [cell growth per unit metabolic heat (g/cal)]

$$Y_H = \frac{Y_{X/S}}{(\Delta H_S - Y_{X/S}\ \Delta H_C)}$$

Typical ΔH_C = 20 to 25 kJ/g dcw

$1/Y_H$; metabolic heat per gram cell produced

Heat generation by growth (cont.)

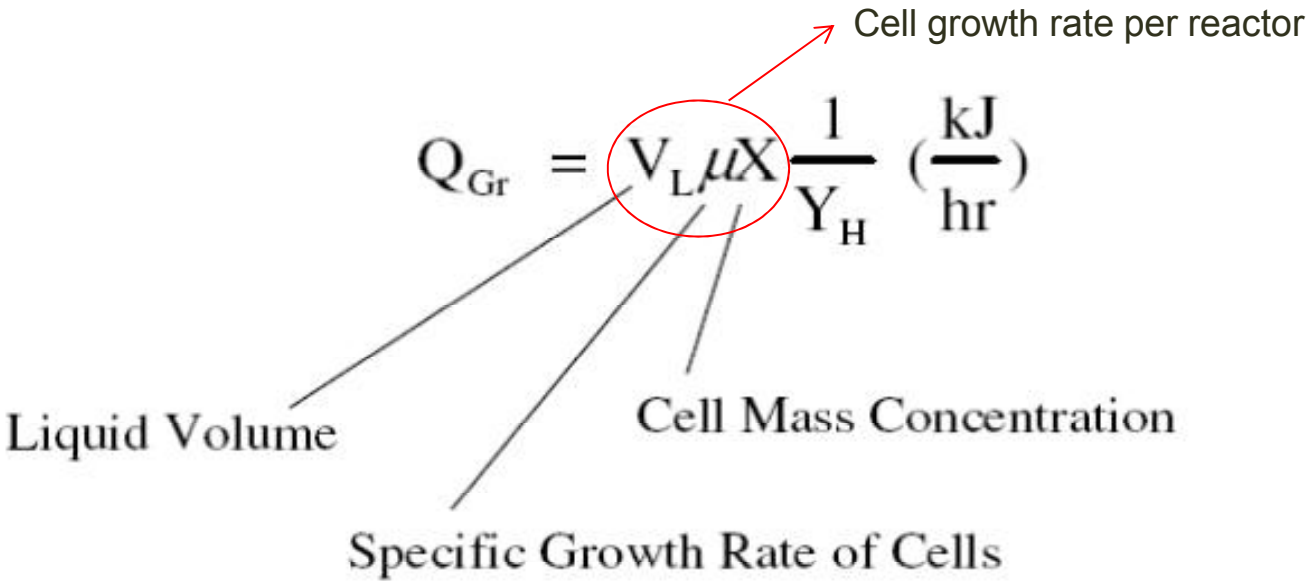
For Substrates:

S	ΔH_S (kJ/g S)	Y_H (g dcw/kJ)
Glucose	15.64	0.072
Methanol	22.68	0.029
Ethanol	29.67	0.043
n-Decane	47.64	0.038
Methane	55.51	0.015

The oxidation state of S has a large effect on 1/ Y_H

Rate of heat generation by cell growth, Q_{Gr}

How can cell growth rate be correlated to heat generation?



Heat can be removed by circulating cooling water through an external jacket around the reactor vessel or through a coiled tube within the reactor.

Modeling cell growth ; Monod equation

Similar to Michaelis-Menten Kinetics

Assumes that a single enzyme system is responsible for the uptake of substrate (S), and that S is limited (growth-dependent variable). This is the most common kinetic model for cell growth.

$$\mu = \frac{\mu_m S}{K_S + S}$$

μ = specific cell growth rate (hr^{-1})

μ_m = maximum specific cell growth rate (hr^{-1})

S = substrate concentration (g/L)

K_S = Saturation constant (g/L) = S when $\mu = 1/2\ \mu_m$.

Batch culture growth model

X(t) = ?

$$\mu = \frac{1}{X}\frac{dX}{dt} = \frac{\mu_m S}{K_S + S} \quad \cdots\cdots (1)$$

We relate changes in S to changes in X through $Y_{X/S}$

$$X - X_o = Y_{X/S}(S_o - S), \text{ or}$$

$$S = S_o + X_o/Y_{X/S} - X/Y_{X/S} \quad \cdots\cdots (2)$$

$Y_{X/S}$ = cell mass yield (g dcw/g S consumed)

X_o, S_o = initial concentrations of cells and substrate

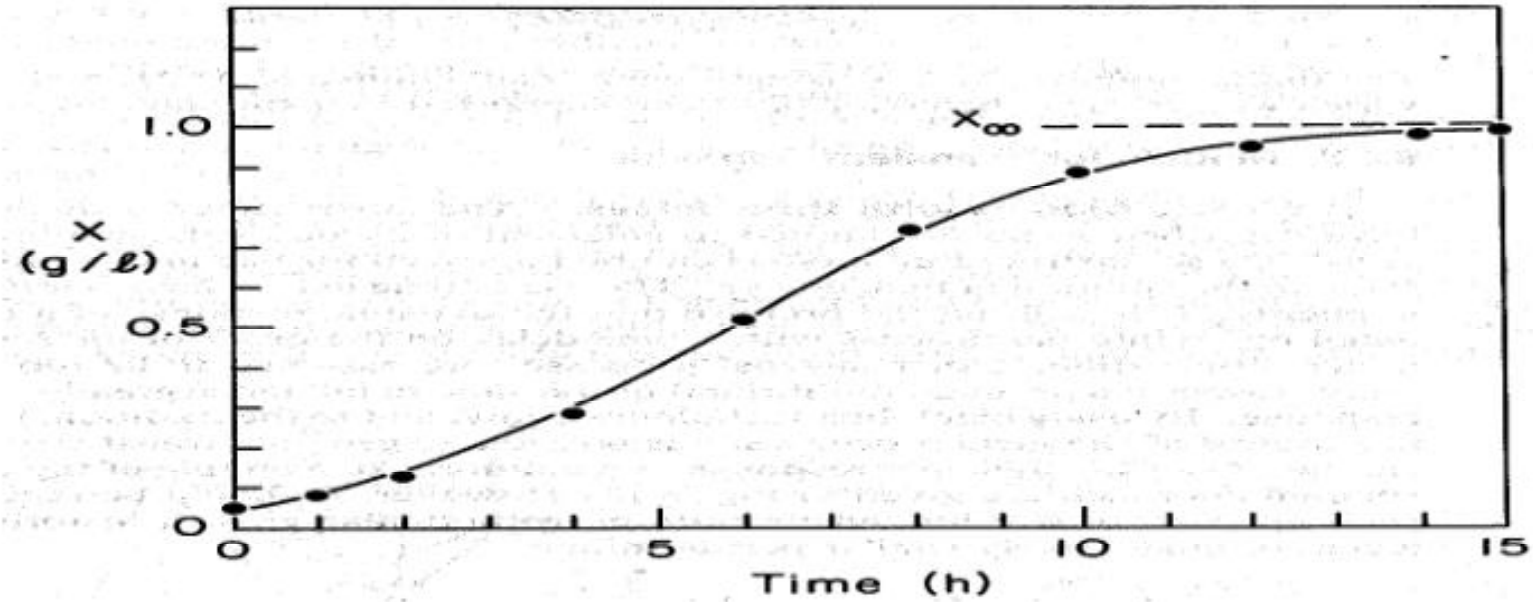
Combine (1) and (2), and rearrange

$$\frac{dX}{dt} = \frac{\mu_m (S_o Y_{X/S} + X_o - X)}{(K_S Y_{X/S} + S_o Y_{X/S} + X_o - X)} X \quad ; \quad \text{at } t = 0,\ X = X_o$$

Batch culture growth model (cont.)

Logistic Equation

$$\frac{(K_S Y_{X/S} + S_o Y_{X/S} + X_o)}{(S_o Y_{X/S} + X_o)} \ln\left(\frac{X}{X_o}\right) - \frac{K_S Y_{X/S}}{(S_o Y_{X/S} + X_o)} \ln\{(S_o Y_{X/S} + X_o - X) S_o Y_{X/S}\} = \mu_m t$$



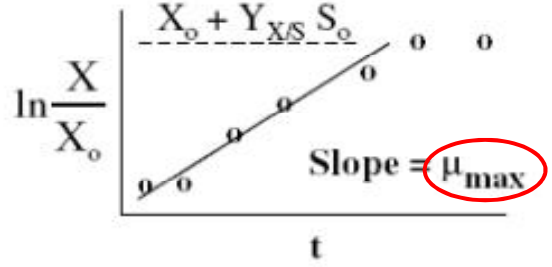
How to determine Monod parameters, K_s and μ_{max}

$$\mu = \frac{1}{X}\frac{dX}{dt}$$

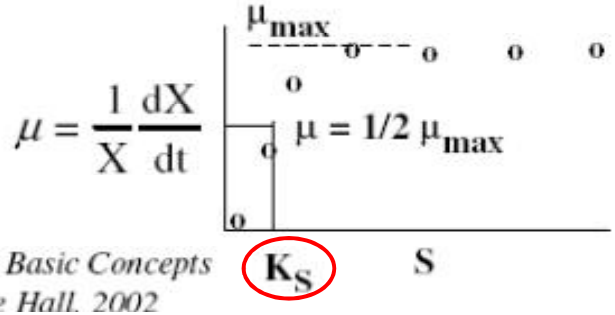
$$\mu\, t = \ln\left(\frac{X}{X_o}\right)$$

K_S is determined differently.

K_S is equal to S when $\mu = 1/2\ \mu_{max}$



$\mu = 1/X\ dX/dt$ needs to be determined from available data, especially data at low S concentrations.



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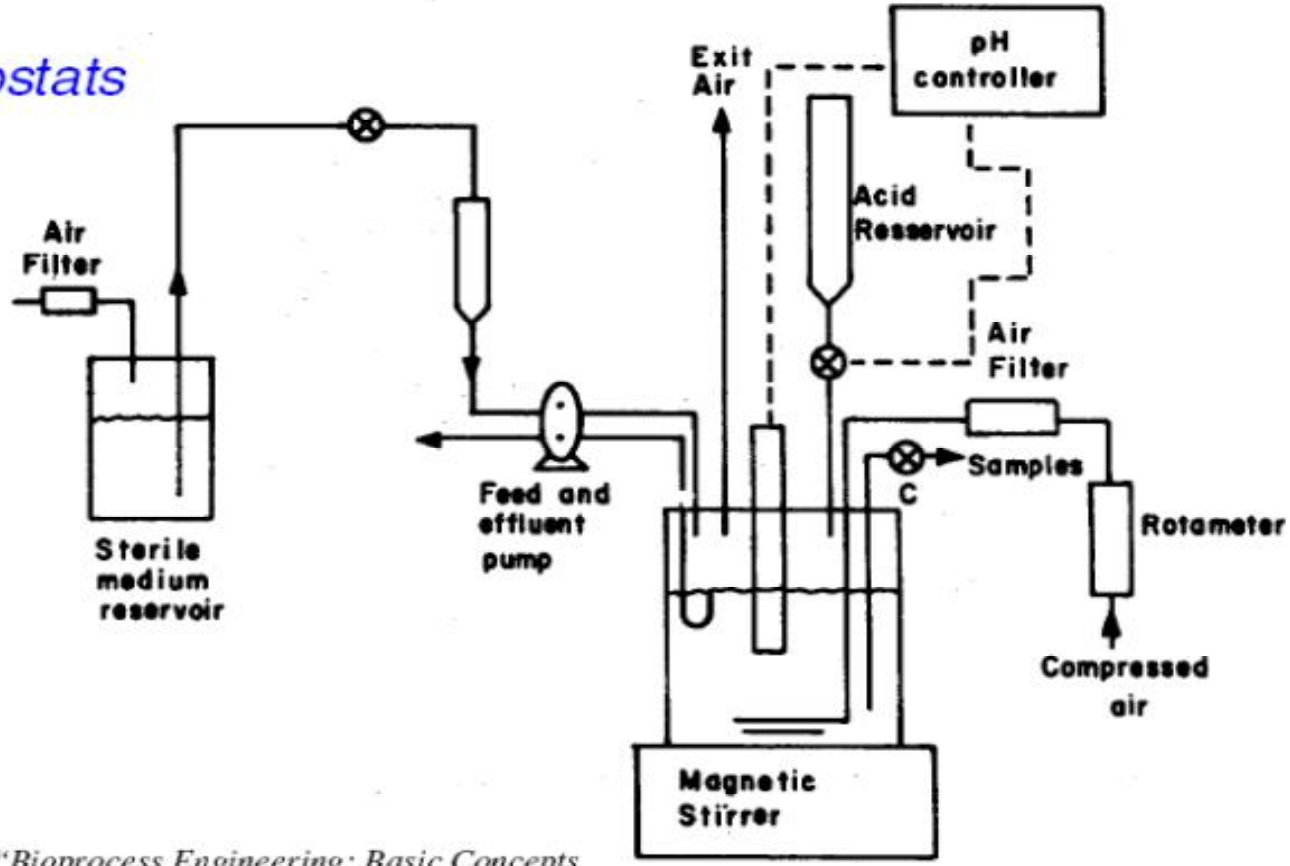
Cell growth in continuous culture

Automated Chemostats

→ control of pH, temp. agitation, dissolved oxygen

→ sterilization required



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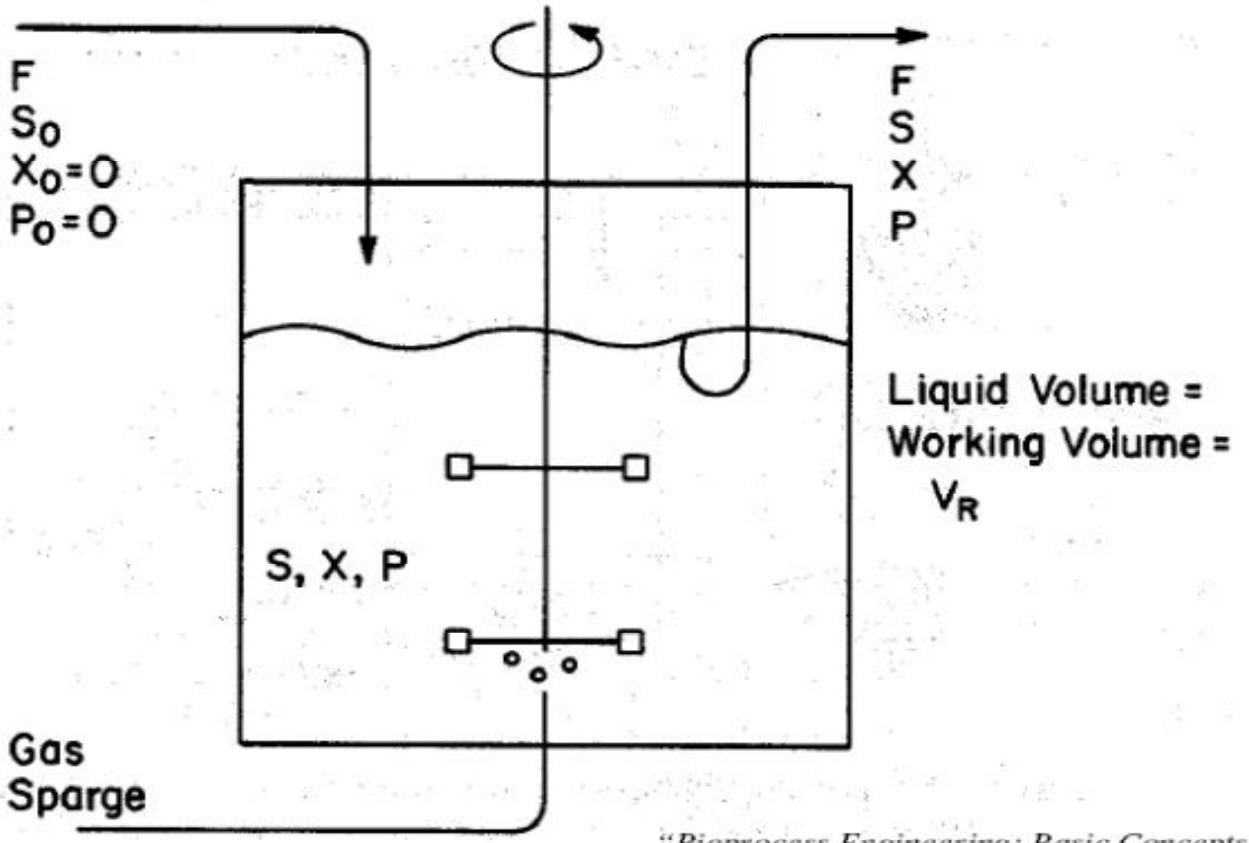
Continuous-stirred tank reactor (CSTR), chemostat



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Cell mass balance in CSTR to get S

$$\begin{bmatrix}\text{mass rate}\\ \text{of cells into}\\ \text{bioreactor}\end{bmatrix} - \begin{bmatrix}\text{mass rate}\\ \text{of cells out}\\ \text{of bioreactor}\end{bmatrix} + \begin{bmatrix}\text{mass rate of cell}\\ \text{growth without}\\ \text{endogenous}\\ \text{metabolism}\end{bmatrix} - \begin{bmatrix}\text{mass rate}\\ \text{of cell loss}\\ \text{by endogenous}\\ \text{metabolism}\end{bmatrix} = \begin{bmatrix}\text{mass rate}\\ \text{of cells}\\ \text{accumulation}\\ \text{in bioreactor}\end{bmatrix}$$

or

$$FX_o - FX + V_R\mu X - V_R k_d X = V_R \frac{dX}{dt}$$

F = in and out volumetric flow rate (L / hr)

X = bioreactor and outlet cell mass concentration (g/L)

X_o = inlet cell mass concentration (g/L) = 0

μ = specific cell growth rate neglecting endogenous metabolism (hr^{-1})

k_d = endogenous cell loss rate constant (hr^{-1})

Steady state and sterile feed

Chemostats are normally operated at steady-state, d/X/dt = 0. Assume a sterile feed ($X_o = 0$), and k_d is so small that is neglected, $k_d = 0$.

The cell mass balance equations becomes,

$$\begin{bmatrix}\text{mass rate}\\ \text{of cells out}\\ \text{of bioreactor}\end{bmatrix} = \begin{bmatrix}\text{mass rate of cell}\\ \text{growth without}\\ \text{endogenous}\\ \text{metabolism}\end{bmatrix}$$

or

$$FX = V_R \mu X$$

$$\frac{F}{V_R} = \mu \quad \text{or} \quad \boxed{D = \mu}$$

where $\frac{F}{V_R} = D$, dilution rate

D [sec^{-1}] ; how many times of rxtor vol. flow per second

Substrate concentration in CSTR when $k_d = 0$

Using the Monod Equation, we can predict the bioreactor and outlet stream concentration of Substrate.

$$\mu = \frac{\mu_{max} S}{K_s + S} = D$$

rearranging, $$\boxed{S = \frac{K_s D}{\mu_{max} - D}}$$

Example: Substrate concentration in the CSTR

The following data were obtained from a batch fermentation.

t (hr)	0	1	2	3	4
X (g/l)	1.00	7.39	54.60	403.43	2980.96

μ was 1 hr^{-1} at a substrate concentration of 2 g/l in the batch fermentation.

In a CSTR with $k_d = 0$, calculate the substrate concentration in the CSTR at D = 1 hr^{-1}.