

## Assignment

**Q.1.** For the manufacture of Swiss cheese, *Lactobacillus casei* is cultivated under anaerobic conditions as a starter culture. The culture produces lactic acid as a by-product of energy metabolism. The organism has the following characteristics:  $Y_{X/S} = 0.23 \text{ Kg/Kg}$ ,  $K_s = 0.15 \text{ Kg/m}^3$ ,  $\mu_{\max} = 0.35 \text{ h}^{-1}$ , and  $m_s = 0.135 \text{ kg/kg h}$ . A stirred-tank reactor is operated in fed batch mode at quasi-steady state with a feed flow rate of  $4 \text{ m}^3/\text{h}$  and feed substrate concentration of  $80 \text{ kg/m}^3$ . After 6 h, the liquid volume is  $40 \text{ m}^3$ .

(a) What was the initial culture volume?

(b) What is the concentration of the substrate at the quasi-steady state?

### Solution:

(a) Given  $F = 4 \text{ m}^3/\text{h}$ ,  $V = 40 \text{ m}^3$ , and  $t = 6 \text{ h}$ .

For a fed batch operation at quasi-steady state (variable volume fed batch), the volume of the reactor at time  $t$  and the initial culture volume  $V_0$  can be given as-

$$V = V_0 + Ft$$

$$V_0 = V - Ft = (40) - (4)(6) = 16 \text{ m}^3$$

(b) Given  $F = 4 \text{ m}^3/\text{h}$ ,  $V = 40 \text{ m}^3$ ,  $K_s = 0.15 \text{ kg/m}^3$ ,  $\mu_{\max} = 0.35 \text{ h}^{-1}$ .

We know that the dilution rate is

$$D = \frac{F}{V} = \frac{4}{40} = 0.10 \text{ h}^{-1}$$

At quasi-steady state,

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

So, the substrate concentration at quasi-steady state can be given as

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

Putting the given values in the above equation, we get

$$S = \frac{0.15(0.10)}{0.35 - 0.10} = 0.06 \text{ kg/m}^3$$

**Q. 2.** The growth of a microorganism on glucose is described by the Monod model. During the growth of the cell, the glucose concentration is  $10 \text{ g/L}$ , and the  $\mu_{\max}$  and  $K_s$  values are  $0.5 \text{ h}^{-1}$  and  $0.1 \text{ g/L}$ , respectively. Find the time required to triple the biomass concentration.

### Solution:

Given  $\mu_{\max} = 0.5 \text{ h}^{-1}$ ,  $K_s = 0.1 \text{ g/L}$ , and  $S = 10 \text{ g/L}$ .

The Monod equation for cell growth is

$$\mu = \frac{\mu_{\max} S}{K_s + S} = \frac{0.5 \times 10}{0.1 + 10} = 0.495 \text{ h}^{-1}$$

If cell death is negligible as compared to growth ( $\mu_g = \mu_{\text{net}}$ ),

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

Rearranging and integrating the above equation, we get

$$\int_{X_0}^X \frac{dX}{X} = \int_0^t \mu dt$$

$$\ln\left(\frac{X}{X_0}\right) = \mu t$$

$$t = \frac{\ln X/X_0}{\mu}$$

Time required to triple cell mass ( $X = 3X_0$ ) is

$$t_{\text{triple}} = \frac{\ln 3}{\mu} = \frac{\ln 3}{0.495}$$

$$t_{\text{triple}} = \frac{\ln 3}{\mu} = 2.22 \text{ h}$$

**Q.3.** *Pseudomonas* sp. has a minimum doubling time of 2.4 h when grown on acetate in a chemostat that follows the Monod model.

Given  $K_s = 1.3 \text{ g/L}$ ,  $Y_{X/S} = 0.46 \text{ g cell / g acetate}$ ,  $S_0 = 38 \text{ g / L}$ .

(a) Find out steady-state  $S$  and  $X$  when  $D = 1/2 D_{\max}$ .

(b) Find the cell mass productivity at  $0.8 D_{\max}$ .

(c) Determine the value of  $D_{\text{washout}}$ .

Solution: It is known that

$$t_{d_{\min}} = \frac{\ln(2)}{\mu_{\max}}$$

$$\mu_{\max} = \frac{\ln(2)}{t_{d_{\min}}} = 0.288 \text{ h}^{-1}$$

$$D_{\max} = \mu_{\max} \left( 1 - \sqrt{\frac{K_s}{K_s + S_0}} \right) = 0.288 \left( 1 - \sqrt{\frac{1.3}{1.3 + 38}} \right)$$

$$= 0.288(1 - 0.1818) = 0.235 \text{ h}^{-1}$$

(a)

$$D = \frac{1}{2} D_{\max} = 0.1178$$

$$S = \frac{K_s D}{\mu_{\max} - D} = \frac{1.3(0.1178)}{0.288 - 0.1178} = 0.899 \frac{\text{g}}{\text{L}}$$

$$X = Y_{X/S} (S_0 - S) = 0.46 (38 - 0.899) = 17 \frac{\text{g}}{\text{L}}$$

(b)  $0.8 D_{\max} = 0.8 \times 0.235 \text{ h}^{-1} = 0.188 \text{ h}^{-1}$

Cell mass productivity is  $XD$ .

$$X = Y_{X/S} (S_0 - S)$$

$$S = \frac{1.3(0.188)}{0.288 - 0.188} = 2.44 \frac{\text{g}}{\text{L}}$$

$$X = 0.46(38 - 2.4) = 16.35 \frac{\text{g}}{\text{L}}$$

Cell mass productivity (at  $0.8 D_{\max}$ ) is

$$XD = 16.35 \times 0.188 = 3.07 \frac{\text{g}}{\text{L h}}$$

(c)

$$D_{\text{washout}} = \frac{\mu_{\max} S_0}{K_s + S_0} = \frac{0.288 \times 38}{1.3 + 38} = 0.278 \text{ h}^{-1}$$

**Q.4.** A  $1 \text{ m}^3$  mixed flow reactor, at the initial substrate concentration of  $S_0 = 500 \text{ g/m}^3$  in the feed, produces  $100 \text{ g/h}$  of yeast cells in the exit stream at two different flow rates mentioned below:

(i) At  $0.5 \text{ m}^3/\text{h}$  of feed for which the steady state  $S = 100 \text{ g/m}^3$

(ii) At  $1 \text{ m}^3/\text{h}$  of feed for which the steady state  $S = 300 \text{ g/m}^3$

The Monod cell growth kinetics follow yeast formation. Compute the following:

(a) The fractional yield of yeast

(b) The kinetic equation for yeast formation

(c) The flow rate for maximum yeast production (Hint: flow rate at  $D_{\max}$ )

(d) The maximum production rate of yeast

Solution:

(a) We know that

Flow rate  $\times$  Concentration of cells = Rate of cell mass production

Therefore, the concentration of yeast is

$$\frac{100 \text{ g/h}}{0.5 \text{ m}^3/\text{h}} = 200 \text{ g/m}^3$$

Case (i): In the case of sterile feed,  $X_{ss} = Y_{X/S} (S_0 - S_{ss})$ .

$$Y_{X/S} = \frac{200 \text{ g/m}^3}{(500 - 100) \text{ g/m}^3} = 0.5$$

(b) From the Monod model under steady state and sterile conditions,

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

Case (i):

$$D = \frac{F}{V} = \frac{0.5 \text{ m}^3/\text{h}}{1 \text{ m}^3} = 0.51/\text{h}$$

So from the Monod equation, we can write

$$0.5 = \frac{\mu_{\max} 100}{K_S + 100}$$

Similarly, in Case (ii)

$$1 = \frac{\mu_{\max} 300}{K_S + 300}$$

$$\mu_{\max} = 2 \frac{1}{\text{h}} \text{ and } K_S = 300 \text{ g/m}^3$$

So, the kinetic equation is:

$$\mu = \frac{2S}{300 + S}$$

(c) We know that

$$D_{\max} = \mu_{\max} \left( 1 - \sqrt{\frac{K_S}{K_S + S_0}} \right)$$

$$D_{\max} = 0.776 \text{ 1/h} = \frac{F}{V}$$

Flow rate for the maximum yeast production is  $0.776 \times 1 = 0.776 \text{ m}^3/\text{h}$ .

(d)

$$S = \frac{K_S D}{(\mu_{\max} - D)} = \frac{300 \times 0.776}{(2 - 0.776)} = 190 \text{ g/m}^3$$

$$X = Y_{X/S} (S_0 - S) = 0.5 (500 - 190) = 155 \text{ g/m}^3$$

Maximum cell mass productivity is  $0.776 \times 155 = 120.28 \text{ g/h m}^3$

**Q. 5.** In a chemostat, the steady-state substrate and biomass concentrations are given in the Table. The initial substrate concentration ( $S_0$ ) is 700 mg/L. Find out  $\mu_{\max}$  and  $K_s$ , the growth yield coefficient,  $Y'_{X/S}$  (growth), and the maintenance coefficient  $m$ .

$D \text{ (h}^{-1}\text{)}$	$S \text{ (mg/L)}$	$X \text{ (mg/L)}$
0.3	45	326
0.25	41	328
0.20	16	340
0.12	8	342
0.08	3.8	344

**Solution:**

The Monod equation is

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

Under steady-state chemostat operation and sterile feed

$$\mu = D$$

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

$$\frac{1}{D} = \frac{K_s}{\mu_{\max}} \frac{1}{S} + \frac{1}{\mu_{\max}}$$

Plotting  $1/D$  versus  $1/S$  yields the value of  $K_s$  and  $\mu_{\max}$ .

$$\mu_{\max} = \frac{1}{2.9} = 0.34 \text{ h}^{-1}$$

$$K_s = 37.53 \times \mu_{\max} = 12.76 \frac{\text{mg}}{\text{L}}$$

According to the Pirt model,

$$\frac{1}{Y_{X/S} \text{ (overall)}} = \frac{1}{Y'_{X/S} \text{ (growth)}} + \frac{m}{\mu}$$

$$\frac{1}{Y_{X/S} \text{ (overall)}} = \frac{1}{Y'_{X/S} \text{ (growth)}} + \frac{m}{D}$$

Plotting  $1/Y_{X/S} \text{ (overall)}$  versus  $1/D$  of Table 6.9 yields the values of  $m$

and  $1/Y'_{X/S} \text{ (growth)}$ .

$D \text{ (h}^{-1}\text{)}$	$1/D \text{ (h)}$	$S \text{ (mg/L)}$	$X \text{ (mg/L)}$	$Y_{X/S(\text{overall})}$	$1/Y_{X/S(\text{overall})}$
0.3	3.33	45	326	0.498	2.008
0.25	4.00	41	328	0.4977	2.009
0.20	5.00	16	340	0.4970	2.012
0.12	8.33	8	342	0.4942	2.023
0.08	12.50	3.8	344	0.4941	2.024

$$Y_{X/S(\text{overall})} = -\frac{dX}{dS} = \frac{X_n - X_0}{S_0 - S_n}$$

For the first sampling point,  $n = 1$

$$Y_{X/S(\text{overall})} = -\frac{dX}{dS} = \frac{X_1 - X_0}{S_0 - S_1} = \frac{326 - 0}{700 - 45} = 0.498$$

From Plot of  $1/Y_{X/S(\text{overall})}$  versus  $1/D$ , we get  $m = 0.0019 \text{ h}^{-1}$  (from slope) and  $Y_{X/S(\text{growth})} = 0.50$  (from intercept of the Y-axis).