UNIVERSITI SAINS MALAYSIA

Second Semester Examination
Academic Session 2007/2008

April 2008

BTT 303/3 – Biochemical Engineering
[Kejuruteraan Biokimia]

Duration: 3 hours
[Masa : 3 jam]

Please ensure that this examination paper contains TEN printed pages before you begin the examination.

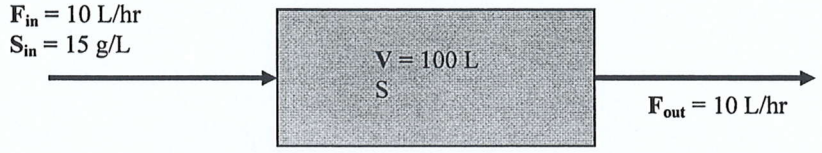
[Sila pastikan bahawa kertas peperiksaan ini mengandungi SEPULUH muka surat yang bercetak sebelum anda memulakan peperiksaan ini.]

Instructions: Answer **FIVE** (5) out of **SIX** (6) questions, in English or Bahasa Malaysia. Each question carries 20 marks.

*[**Arahan:** Jawab **LIMA** (5) daripada **ENAM** (6) soalan yang diberikan dalam Bahasa Inggeris atau Bahasa Malaysia. Tiap-tiap soalan bernilai 20 markah.]*

...2/-

1. A 100L fermenter is initially filled with sterile carbon-free medium. A 15 g/L sterile glucose solution is then fed into the fermenter at a rate of 10 L/hour. An outflow of 10 L/hour is also started.



[a] Write the glucose mass balance in the fermenter for the conditions below:

[i] The fermenter has not reached steady-state.

(5 marks)

[ii] The fermenter is in steady-state.

(5 marks)

[iii] The fermenter has been inoculated with a yeast cell suspension, x_0, at 0.5 g/L and has reached steady-state. Simplify the equation so that it does not contain unknown biomass term (x).

(10 marks)

2. [a] You have been assigned to solve the problem of waste water effluent from the plant where you work. Suggest the appropriate unit operations required in addressing and rectifying this problem. Justify your suggestions.

(10 marks)

[b] Microbial product synthesis can be classified into several categories, based on the kinetic patterns of growth and product formation. With the help of diagrams, list these categories and the corresponding equation for the specific rate of product formation, q_p. Give examples for each case.

(10 marks)

3. A strain of mold is grown in a batch culture containing glucose as a substrate. The cell dry weight and glucose concentration are measured at each sampling point. The data obtained are as follows.

Time (h)	Cell concentration (g/L)	Glucose concentration (g/L)
0	0.75	60.0
9	1.47	58.2
16	3.06	54.2
23	6.3	46.1
30	13.2	28.9
34	19.8	12.4
36	22.5	5.6
40	24.6	0.4

[a] Plot :

[i] The fungus growth curve.

(3 marks)

[ii] the glucose consumption curve.

(3 marks)

[b] From the plots above, estimate :

[i] The maximum specific growth rate.

(3 marks)

[ii] The substrate yield.

(3 marks)

[c] Estimate the maximum cell concentration that can be achieved if the inoculum size was the same but the initial glucose concentration was 100 g/L (instead of 60 g/L glucose in the table above).

(4 marks)

[d] Based on the data above, is the fungus showing dispersed filamentous growth or pelleted growth? Justify your answer.

(4 marks)

4.

V ([E_0] = 0.015 g/L) g/L.min	[S] g/L	V ([E_0] = 0.0075 g/L) g/L.min
1.14	20.0	0.67
0.87	10.0	0.51
0.7	6.7	0.41
0.59	5.0	0.34
0.5	4.0	0.29
0.44	3.3	
0.39	2.9	
0.35	2.5	

The table above shows enzymatic reaction data obtained using two different initial enzyme concentrations.

Based on the data,

[a] Plot the appropriate graph.

[b] Using the graph, estimate:

[i] K_m

[ii] V_{max} for $[E_0]$ = 0.015 g/L

[iii] V_{max} for $[E_0]$ = 0.0075 g/L

[c] Estimate the value of k_2

(15 marks)

[d] Is the V_{max} value affected by the initial enzyme concentration? Why?

(5 marks)

5. [a] Describe the following ideal reactors, listing their general characteristics and the corresponding mass-balance equation.

[i] Continuous Stirred Tank Reactor.

(5 marks)

[ii] Plug Flow Tubular Reactor.

(5 marks)

[b] Compare and contrast the following terms

[i] Open system, closed system.

(5 marks)

[ii] Steady state, unsteady (transient) state.

(5 marks)

6. Interleukin-3 (IL-3) is a type of biological signal (cytokine) that can improve the body's natural response to disease as part of the immune system. You have established the fermentation protocol for producing IL-3 in recombinant *Aspergillus awamori* shake flask cultures. You are interested in increasing the production volume.

[a] Describe two properties that can be used as the basis for scale-up.

(4 marks)

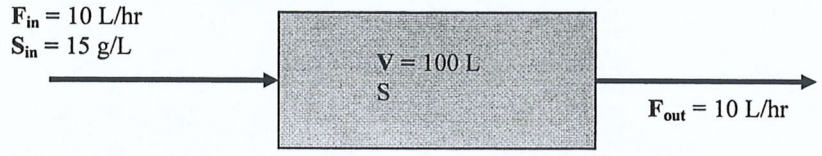
[b] Elaborate on the issues of scaling-up a fermentation process.

(6 marks)

[c] IL-3 fermentation is an aerobic process, achieved in an aerated stirred tank bioreactor. Discuss the factors that can influence the value of the oxygen transfer coefficient in the bioreactor.

(10 marks)

1. Fermenter berisipadu 100 L pada mulanya dipenuhkan dengan medium tanpa karbon yang steril. Kemudian, suapan glukosa steril berkepekatan 15 g/L dimasukkan dengan kadar 10 L/jam. Aliran keluar juga dimulakan dengan kadar 10 L/jam.



Tulis persamaan imbangan jisim glukosa bagi fermenter ini untuk keadaan-keadaan berikut:

[i] Fermenter belum mencapai keadaan mantap.

(5 markah)

[ii] Fermenter berada dalam keadaan mantap.

(5 markah)

[iii] Fermenter telah diinokulatkan dengan ampaian sel yis pada 0.5 g/L dan telah mencapai keadaan mantap. Persamaan ini perlu diringkaskan supaya tidak mengandungi sebutan kepekatan biojisim yang tidak diketahui (x).

(10 markah)

2. [a] Anda telah ditugaskan untuk menyelesaikan masalah sisa air buangan di kilang tempat anda bekerja. Cadangkan unit operasi yang sesuai dalam menangani dan mengatasi masalah ini. Berikan alasan untuk cadangan anda.

(10 markah)

[b] Sintesis produk mikrob boleh dikelaskan kepada beberapa kategori, berdasarkan corak kinetik pertumbuhan dan penghasilan produk. Dengan bantuan gambar rajah, senaraikan kategori ini dan persamaan untuk kadar penghasilan produk spesifik, q_p, yang berkaitan. Berikan contoh untuk setiap kes.

(10 markah)

3. Sejenis kulat ditumbuhkan di dalam kultur kelompok yang mengandungi glukosa sebagai substrat. Berat kering biojisim dan kepekatan glukosa diukur pada setiap masa persampelan. Data yang diperoleh adalah seperti berikut:

Masa (jam)	Kepekatan sel (g/L)	Kepekatan glukosa (g/L)
0	0.75	60.0
9	1.47	58.2
16	3.06	54.2
23	6.3	46.1
30	13.2	28.9
34	19.8	12.4
36	22.5	5.6
40	24.6	0.4

[a] Plotkan :

[i] Keluk pertumbuhan kulat.

(3 markah)

[ii] Keluk penggunaan glukosa

(3 markah)

[b] Daripada plot di atas, anggarkan :

[i] Kadar pertumbuhan spesifik maksimum.

(3 markah)

[ii] Pekali hasil bagi substrat

(3 markah)

[c] Anggarkan kepekatan sel maksimum yang boleh dicapai sekiranya saiz inokulum adalah sama tetapi kepekatan awal glukosa ialah 100 g/L (bukan 60 g/L seperti di dalam jadual di atas).

(4 markah)

[d] Berdasarkan kepada maklumat yang diperoleh, adakah kulat ini menunjukkan pertumbuhan berfilamen atau gumpalan? Nyatakan alasan anda.

(4 markah)

4. Jadual di atas menunjukkan data tindak balas enzim yang diperoleh untuk dua kepekatan enzim awal yang berbeza.

Berdasarkan kepada data,

[a] Plotkan graf yang sesuai.

[b] Daripada plot, anggarkan:

[i] K_m

[ii] V_{maks} untuk $[E_0] = 0.015$ g/L

[iii] V_{maks} untuk $[E_0] = 0.0075$ g/L

[c] Anggarkan nilai k_2

(15 markah)

[d] Adakah nilai V_{maks} dipengaruhi oleh kepekatan enzim awal? Kenapa?

(5 markah)

5. [a] Terangkan reaktor ideal berikut, senaraikan ciri-ciri am serta persamaan imbangan jisim yang berkaitan.

[i] Reaktor Tangki Teraduk Selanjar.

(5 markah)

[ii] Reaktor Tiub Aliran Palam.

(5 markah)

[b] Bandingkan dan bezakan istilah-istilah berikut:

[i] Sistem tertutup, sistem terbuka.

(5 markah)

[ii] Keadaan mantap, keadaan tak mantap (peralihan).

(5 markah)

6. Interleukin-3 (IL-3) ialah sejenis isyarat biologi (sitokin) yang boleh meningkatkan gerak balas asli badan terhadap penyakit sebagai sebahagian daripada sistem imun. Anda telah memantapkan protokol fermentasi bagi menghasilkan IL-3 dalam kultur kelalang goncangan *Aspergillus awamori* rekombinan. Anda berminat meningkatkan isipadu penghasilan.

[a] Terangkan dua sifat yang boleh digunakan sebagai asas untuk peningkatan skala.

(4 markah)

[b] Huraikan isu peningkatan skala sesuatu proses fermentasi.

(6 markah)

[c] Fermentasi IL-3 ialah satu proses aerob yang dicapai di dalam bioreaktor tangki teraduk berudara. Bincangkan faktor-faktor yang boleh mempengaruhi nilai koefisyen pemindahan oksigen di dalam bioreaktor.

(10 markah)

$$Y_{x/s} = -\frac{\Delta x}{\Delta S} = \frac{(x - x_0)}{(S_0 - S)}$$

$$Y_{p/s} = -\frac{\Delta P}{\Delta S} = \frac{(P - P_0)}{(S_0 - S)}$$

$$\frac{1}{Y_m} = \frac{m}{\mu} + \frac{1}{Y_G}$$

$$q_p = \frac{1}{X}\frac{dP}{dt} = \mu Y_{p/x}$$

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

$$\mu = \frac{\mu_{max} S}{K_{S0} S_0 + S}$$

$$\mu = \frac{\mu_{max} S}{K_{S1} + K_{S0} S_0 + S}$$

$$S_{max} = \sqrt{K'_m K_{SI}}$$

$$x = x_0 e^{\mu t}$$

$$\frac{\ln 2}{\mu} = \frac{0.693}{\mu} = t_d$$

$$\frac{dc_i}{d\tau} = r_i$$

$$v = \frac{v_{max} S}{K_M + S}$$

$$V_{max} - \frac{S_0 - S}{t} = \frac{K_m}{t} \ln \frac{S_0}{S}$$

$$\frac{1}{v} = \frac{1}{V_{max}} + \frac{K_m}{V_{max}} \frac{1}{[S]}$$

$$v = V_{max} - K_m \frac{v}{[S]}$$

$$\frac{[S]}{v} = \frac{K_m}{V_{max}} + \frac{1}{V_{max}}[S]$$

$$v = -\frac{d[S]}{dt} = \frac{V_{max}[S]^n}{K_m^{II} + [S]^n}$$

$$v = \frac{V_{max}[S]}{K_m\left(1 + \frac{[I]}{K_i}\right) + [S]}$$

$$v = \frac{\frac{V_{max}}{\left(1 + \frac{[I]}{K_i}\right)}[S]}{\frac{K'_m}{\left(1 + \frac{[I]}{K_i}\right)} + [S]}$$