UNIVERSITI SAINS MALAYSIA

First Semester Examination
2016/2017 Academic Session

December 2016 / January 2017

EKC 271 – Biotechnology for Engineers
[Bioteknologi untuk Jurutera]

Duration : 3 hours
[Masa : 3 jam]

Please check that this examination paper consists of SEVEN pages of printed material and ONE page of Appendix before you begin the examination.

[Sila pastikan bahawa kertas peperiksaan ini mengandungi TUJUH muka surat yang bercetak dan SATU muka surat Lampiran sebelum anda memulakan peperiksaan ini.]

Instruction: Answer **ALL** (4) questions.

*[**Arahan:** Jawab **SEMUA** (4) soalan.]*

In the event of any discrepancies, the English version shall be used.

[Sekiranya terdapat sebarang percanggahan pada soalan peperiksaan, versi Bahasa Inggeris hendaklah diguna pakai.]

Answer ALL questions.

1. [a] [i] What type of enzyme inhibition does Figure Q.1.[a] indicate? Give 2 reasons.

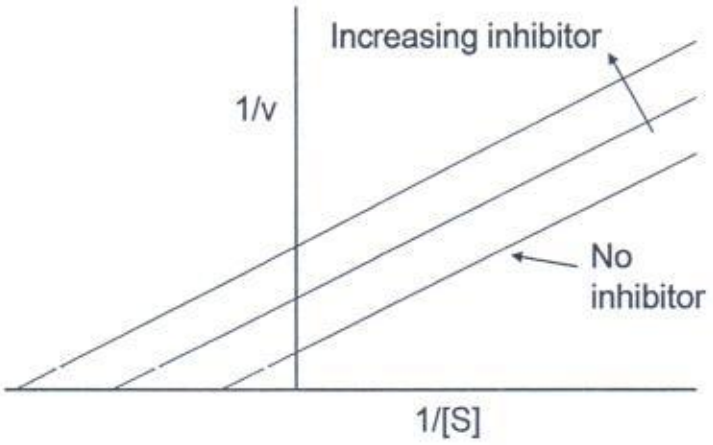


Figure Q.1.[a]

[3 marks]

[ii] What is the chemical similarities or differences between substrate and the inhibitor?

[2 marks]

[iii] Draw the reaction scheme for the above inhibition.

[5 marks]

[b] Sucrase catalyses the hydrolysis of sucrose to produce glucose and fructose. Experiments are carried out to determine the kinetic parameters for the enzyme. Initial rate data are given in Table Q.1.[b].

Table Q.1.[b]

Sucrose concentration (mol $L^{-1} \times 10^{-2}$)	Initial reaction velocity (mol $L^{-1}min^{-1} \times 10^{3}$)
2.50	1.94
2.27	1.91
1.84	1.85
1.35	1.80
1.25	1.78
0.73	1.46
0.46	1.17
0.204	0.779

[i] Use least square method to calculate the V_{max} and K_M.

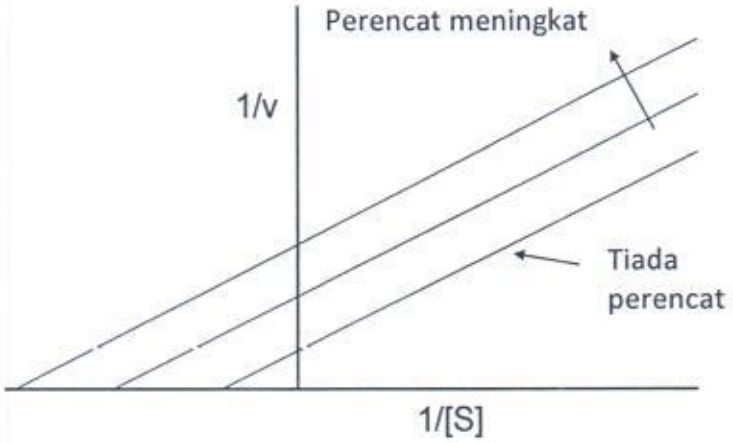
[12 marks]

[ii] Describe what will happen to the K_M and V_{max} values when an uncompetitive inhibitor is added to the reaction mixture.

[3 marks]

Jawab SEMUA soalan.

1. [a] [i] *Apakah jenis perencatan enzim yang ditunjukkan oleh Rajah S.1.[a]? Berikan 2 sebab.*



Rajah S.1.[a]

[3 markah]

[ii] *Apakah persamaan atau perbezaan kimia antara substrat dan perencat?*

[2 markah]

[iii] *Lukiskan skema tindak balas untuk perencatan di atas.*

[5 markah]

[b] *Sukrase memangkinkan hidrolisis sukrosa untuk menghasilkan glukosa dan fruktosa. Eksperimen dijalankan untuk menentukan parameter kinetik bagi enzim tersebut. Data kadar awalan diberikan dalam Jadual S.1.[b].*

Jadual S.1.[b]

Kepekatan Sukrosa (mol $L^{-1} \times 10^{-2}$)	*Halaju Tindak balas Awalan (mol $L^{-1}min^{-1} \times 10^{3}$)*
2.50	*1.94*
2.27	*1.91*
1.84	*1.85*
1.35	*1.80*
1.25	*1.78*
0.73	*1.46*
0.46	*1.17*
0.204	*0.779*

[i] *Gunakan kaedah kuasa dua terkecil untuk mengira* V_{max} *dan* K_M.

[12 markah]

[ii] *Gambarkan apa yang akan terjadi pada nilai* K_M *dan* V_{max} *apabila suatu perencat tak-kompetitif ditambahkan ke dalam campuran tindak balas.*

[3 markah]

2. [a] [i] Name the two types of nucleic acid in cells and list down their differences.

[4 marks]

[ii] How can we differentiate *α-D-glucose* from *β-D-glucose*?

[4 marks]

[iii] Explain why fructose is capable of acting as a reducing agent.

[4 marks]

[b] [i] Distinguish between anabolic and catabolic processes.

[3 marks]

[ii] Yogurt is produced through the fermentation of milk by *Lactobacillus bulgaricus* and *Streptococcus thermophiles*. Describe the metabolic process of these bacteria for the production of yogurt.

[10 marks]

3. [a] [i] A laboratory-scale bioreactor is filled with medium and then sterilized in a steam autoclave. Explain about the sterilization process and sketch the time-temperature profile.

[6 marks]

[ii] Name the best technique for air sterilization for aerobic fermentation of *Saccharomyces cerevisiae*. Discuss this technique in detail.

[7 marks]

[b] A liquid medium initially contains contaminating *Bacillus stearothermophilus* spores at a concentration of $9.2 \times 10^{11} m^{-3}$. The medium is sterilized thermally at 125°C, and spores density is recorded. The data are given in Table Q.3.[b].

Table: Q.3.[b]

Time (min)	0	5	10	20	30	35
Spore density (m^{-3})	9.2×10^{11}	2×10^{10}	3.32×10^{9}	2.3×10^{5}	115	3

[i] Plot the relevant graph and find the thermal death kinetic rate constant in s^{-1}.

[4 marks]

[ii] Calculate the inactivation factor (N_0/N_t) after 40 min.

[8 marks]

4. [a] Compare prokaryotes with eukaryotes in terms of internal structure and function.

[4 marks]

[b] You were asked to develop a medium for the production of an antibiotic from a newly isolated fungus. The antibiotic was to be produced in an industrial scale and it is relatively inexpensive. The fungus is isolated from soil and the nutritional requirements for rapid growth are uncertain. In this case, would you try to develop a defined or a complex medium? Give your justifications.

[4 marks]

...5/-

2. [a] [i] *Namakan kedua-dua jenis asid nukleik di dalam sel-sel dan senaraikan perbezaannya.*

[4 markah]

[ii] *Bagaimana kita boleh membezakan α-D-glukosa daripada β-D-glukosa?*

[4 markah]

[iii] *Terangkan mengapa fruktosa mampu bertindak sebagai agen penurunan?*

[4 markah]

[b] [i] *Bezakan antara proses anabolik dan proses katabolik.*

[3 markah]

[ii] *Yogurt adalah dihasilkan melalui penapaian susu oleh Lactobacillus bulgaricus dan Streptococcus thermophiles. Jelaskan proses metabolisma bakteria-bakteria ini untuk pengeluaran yogurt.*

[10 markah]

3. [a] [i] *Bioreaktor berskala makmal dipenuhi dengan medium dan kemudian disterilkan dalam autoklaf stim. Terangkan mengenai proses pensterilan dan lakarkan profil masa-suhu.*

[6 markah]

[ii] *Namakan teknik terbaik untuk pensterilan udara untuk penapaian aerobik Saccharomyces cerevisiae. Bincangkan teknik ini secara terperinci.*

[7 markah]

[b] *Medium cecair pada mulanya mengandungi pencemaran spora-spora Bacillus stearothermophilus pada kepekatan* $9.2 \times 10^{11} m^{-3}$. *Medium ini disterilkan secara termal pada 125°C, dan ketumpatan spora-spora direkodkan. Data diberikan dalam Jadual S.3.[b].*

Jadual: S.3.[b]

Masa (min)	*0*	*5*	*10*	*20*	*30*	*35*
Ketumpatan spora (m^{-3})	9.2×10^{11}	2×10^{10}	3.32×10^{9}	2.3×10^{5}	*115*	*3*

[i] *Plot graf yang berkaitan dan cari pemalar kadar kinetik kematian termal dalam* s^{-1}

[4 markah]

[ii] *Kira faktor pentakaktifan* (N_0 / N_t) *selepas 40 min.*

[8 markah]

4. [a] *Bandingkan prokariot dengan eukariot dari segi struktur dalaman dan fungsi.*

[4 markah]

[b] *Anda telah diminta untuk membangunkan medium untuk pengeluaran antibiotik dari kulat baru yang dipencil. Antibiotik itu akan dihasilkan dalam skala industri dan ia adalah agak murah. Kulat tersebut dipencil dari tanah dan keperluan nutrisi untuk pertumbuhan pesat tidak dikenalpasti. Dalam kes ini, adakah anda akan cuba untuk membangunkan media yang ditakrif atau medium kompleks. Berikan justifikasi anda.*

[4 markah]

...6/-

[c] A batch fermentation of *Lactobacillus* sp. growing on lactose gave the results shown in Table Q.4.[c].

Table Q.4.[c]

Time (h)	X (mg/l)	S (g/l)
0	0.20	9.23
2	0.21	9.21
4	0.31	9.07
8	0.98	8.03
10	1.77	6.80
12	3.20	4.60
14	5.60	0.92
16	6.15	0.087
18	6.20	0.00

Calculate;

[i] Yield on substrate, $Y_{X/S}$

[2 marks]

[ii] Mass doubling time, t_d

[2 marks]

[iii] Specific growth rate (μ_{net}) at t=10 h

[2 marks]

[d] The following data are obtained from the oxidation of herbicides present in wastewater by a mix culture of microorganism in a continuously operation aeration tank.

D (h^{-1})	S (mg/l)	X (mg/l)
0.05	15	162
0.11	25	210
0.24	50	250
0.39	100	235
0.52	140	220
0.70	180	205
0.82	240	170

Assuming the herbicide concentration in the feed wastewater stream as S_0=500 mg/L, determine the following;

[i] $Y_{X/S}^{M}$

[2 marks]

[ii] k_d

[2 marks]

[iii] μ_m

[2 marks]

[iv] K_S

[2 marks]

[v] If your main purpose is to reduce the herbicide concentration present in the wastewater to a concentration of 200 mg/l, what would be the corresponding doubling time of the microorganism in the wastewater?

[3 marks]

...7/-

[c] *Penapaian kelompok Lactobacillus sp. tumbuh menggunakan laktosa memberikan keputusan yang ditunjukkan dalam Jadual S.4.[c].*

Jadual S.4.[c]

Masa (j)	X (mg/l)	S (g/l)
0	0.20	9.23
2	0.21	9.21
4	0.31	9.07
8	0.98	8.03
10	1.77	6.80
12	3.20	4.60
14	5.60	0.92
16	6.15	0.087
18	6.20	0.00

Kirakan;

[i] *Hasil dari substrat,* $Y_{X/S}$

[2 markah]

[ii] *Masa penggandaan dua jisim,* t_d

[2 markah]

[iii] *Kadar pertumbuhan spesifik* (μ_{net}) *pada t=10 j*

[2 markah]

[d] *Data berikut diperolehi daripada pengoksidaan herbasid di dalam air sisa oleh campuran kultur mikroorganisma di dalam tangki operasi pengudaraan berterusan.*

D (j^{-1})	S (mg/l)	X (mg/l)
0.05	15	162
0.11	25	210
0.24	50	250
0.39	100	235
0.52	140	220
0.70	180	205
0.82	240	170

Dengan mengandaikan kepekatan racun herba dalam aliran suapan air sisa sebagai S_0 = 500 mg / L, *tentukan yang berikut*

[i] $Y^M_{X/S}$ [2 markah]

[ii] k_d [2 markah]

[iii] μ_m [2 markah]

[iv] K_S [2 markah]

[v] *Jika tujuan utama anda adalah untuk mengurangkan herbasid di dalam air sisa kepada kepekatan 200 mg/l, apakah masa penggandaan dua jisim yang sepadan bagi mikroorganisma dalam air sisa?*

[3 markah]

-oooOooo-

Appendix

$$-\frac{\mathrm{d}n}{\mathrm{d}t} = k_\mathrm{d} n$$

$$k_\mathrm{d} = k_{\mathrm{d}0} \mathrm{e}^{-E_\mathrm{a}/RT}$$

$$\ln\frac{n}{n_0} = -k_{\mathrm{d}0}\int_0^t \mathrm{e}^{-E_\mathrm{a}/RT}\mathrm{d}t$$

$$\ln\frac{n_\mathrm{f}}{n_0} = \ln\frac{n_\mathrm{heat}}{n_0} + \ln\frac{n_\mathrm{hold}}{n_\mathrm{heat}} + \ln\frac{n_\mathrm{f}}{n_\mathrm{hold}}$$

$$\ln\frac{n_0}{n_\mathrm{f}} = k_{\mathrm{d}0}\mathrm{e}^{-E_\mathrm{a}/RT}\tau_\mathrm{hold}$$

$$\tau_\mathrm{hold} = \ln\frac{n_0}{n}/k_\mathrm{d}$$

$$(\mathrm{Pe}) = (vL/E_\mathrm{Dz})$$

slope of regression line $$m = \frac{\Sigma(x_i - \bar{x})(y_i - \bar{y})}{\Sigma(x_i - \bar{x})^2}$$

$$b = \bar{y} - m\bar{x}$$

Easier Form of Least Squares Equations $$m = \frac{\Sigma x_i y_i - [(\Sigma x_i \Sigma y_i)/n]}{\Sigma x_i^2 - [(\Sigma x_i)^2/n]}$$

- n is the number of data points

Monod Equation: $\mu_g = \frac{\mu_{max} S}{K_s + S}$

Biomass balance: $\frac{dX}{dt} = DX_o + (\mu_g - k_d - D)K$ with D=F/V_R

Material balance on limiting substrate: $FS_o - FS + V_r \mu_g X \frac{1}{Y^M_{X/S}} - V_r q_p X \frac{1}{Y_{P/S}} = V_r \frac{dS}{dt}$