
UNIVERSITI SAINS MALAYSIA

First Semester Examination
2010/2011 Academic Session

November 2010

IBG 303 – DNA and Metabolite Technology
[Teknologi DNA dan Metabolit]

Duration: 3 hours
[Masa: 3 jam]

Please check that this examination paper consists of NINE pages of printed material before you begin the examination.

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

Instructions: Answer all FIVE questions. You may answer the questions either in Bahasa Malaysia or in English.

Arahan: *Jawab semua LIMA soalan. Anda dibenarkan menjawab soalan sama ada dalam Bahasa Malaysia atau Bahasa Inggeris.]*

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.]

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Answer ALL questions.

1. Answer all parts of this question.

- (a) Explain the function of a promoter in a bacterial vector. (2 marks)
- (b) A group of genes under the control of a single promoter is called an operon. Illustrate the entire lac operon (4 marks)
- (c) Explain the function of the lactose repressor control mechanism when:-
 - (i) Lactose concentration is present (7 marks)
 - (ii) Lactose concentration is absent (7 marks)

2. Answer all parts of this question.

- (a) There are three different types of DNA preparation. Name the three types of DNA preparation and briefly explain them. (12 marks)
- (b) Describe the DNA purification process using ion exchange chromatography. (8 marks)

3. Answer all parts of this question.

- (a) Describe the three main stages in the central dogma of molecular biology. (3 marks)
- (b) Describe the functions of the following RNA molecules.
 - (i) mRNA
 - (ii) tRNA
 - (iii) rRNA(3 marks)

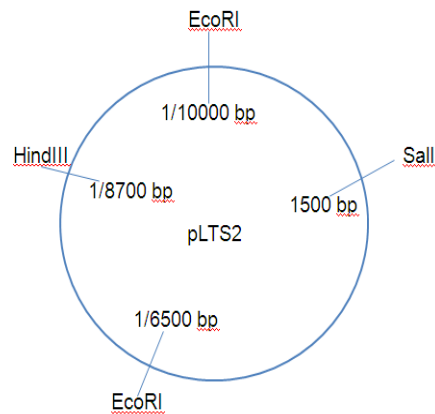
- (c) The sequence of the DNA template provided to you is as follows:

5'-TAC CCC ATT CAT TAC CCT ATC-3'

- (i) What is the sequence of the complementary DNA strand?
 (ii) What is the sequence of the mRNA strand?

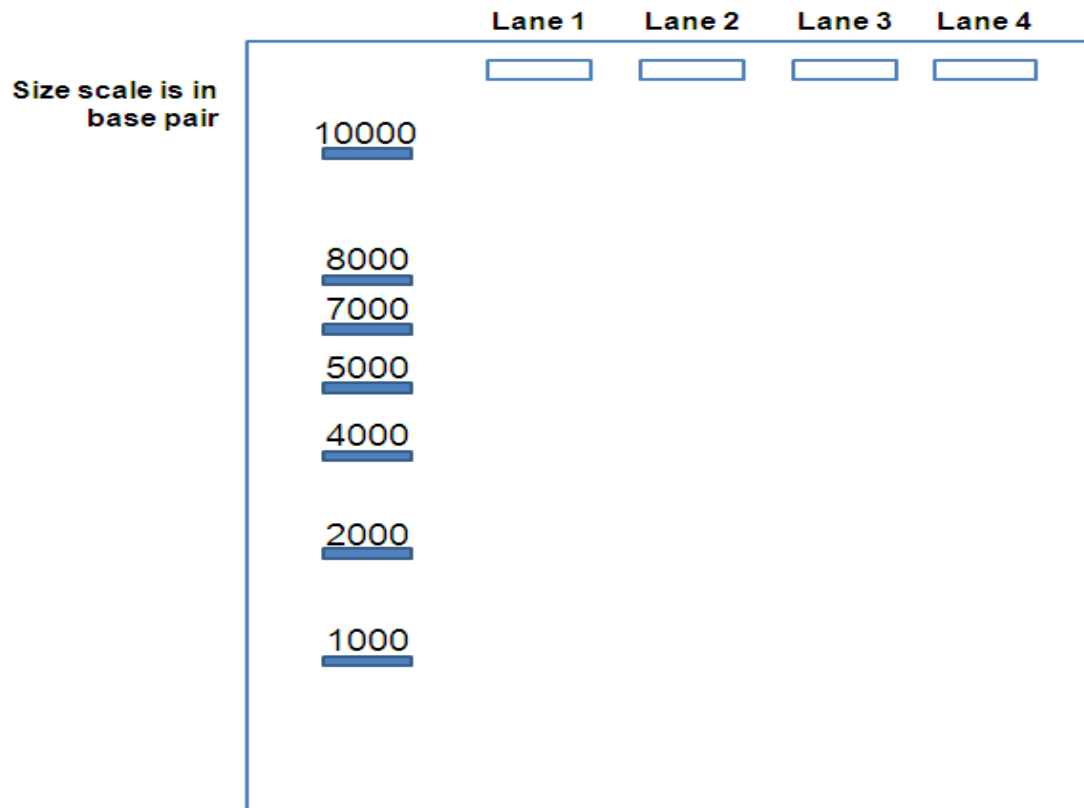
(2 marks)

- (d) Using the vector map provided below, complete the following electrophoresis gel below with the selected digestions.



- (i) Lane 1: with the digestion using only EcoRI
 (ii) Lane 2: with the digestion using Sall and HindIII
 (iii) Lane 3: with the digestion using Sall and EcoRI
 (iv) Lane 4: with the digestion using all the enzymes

(12 marks)



4. Answer all parts of this question.

(a) Describe the three main steps (denaturation, annealing and extension) in polymerase chain reaction (PCR).

(9 marks)

(b) Describe two important parameters to consider when conducting a PCR experiment.

(6 marks)

(c) Calculate the number of DNA molecules present in a 1 ng plasmid with the size of 5 kb. The average size of a nucleotide is 649 daltons.

(5 marks)

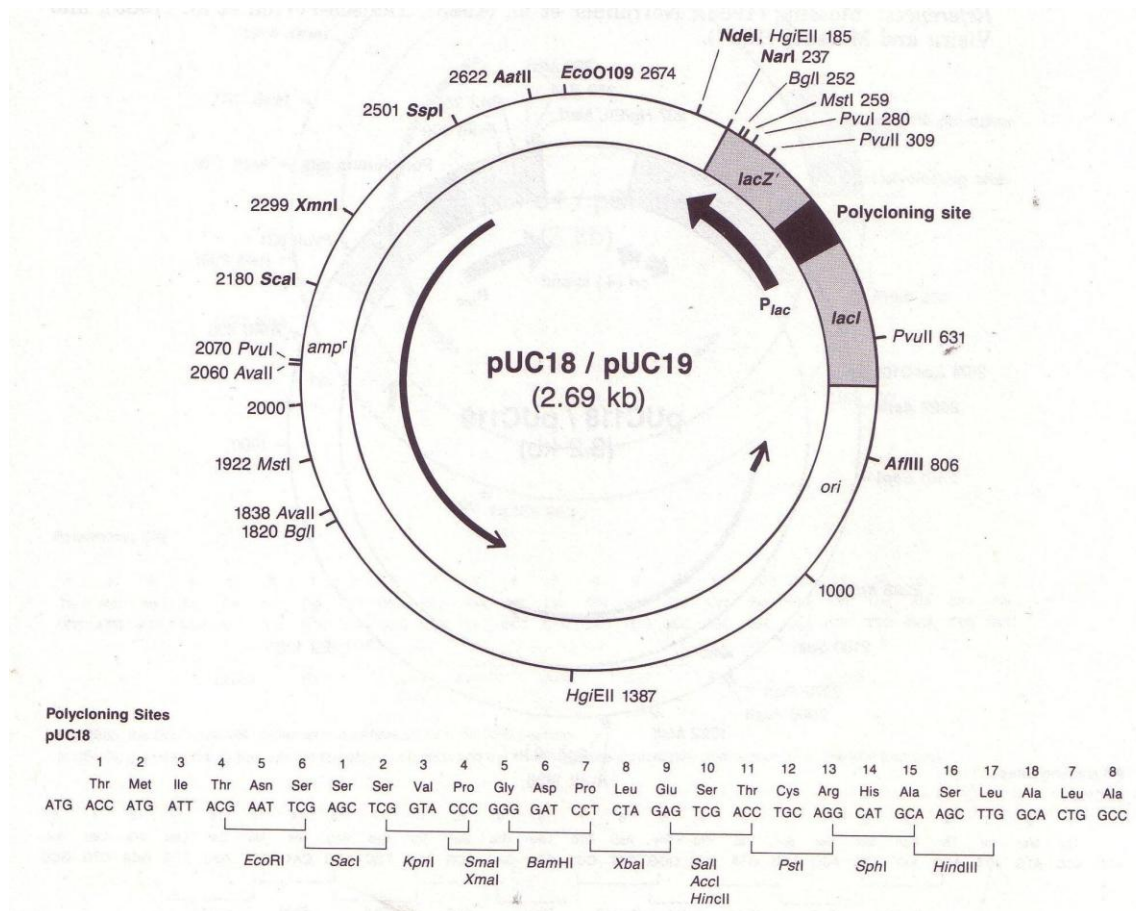
5. Answer all parts of this question.

(a) Explain the significance of the following reagents as applicable to the bacterial DNA extraction procedure: lysozyme, RNase and absolute ethanol

(6 marks)

...5/-

- (b) Prepare a reaction mixture for a restriction digest containing components from the following stock solutions: plasmid DNA (2 mg/ml), reaction buffer (10x), and BamH1 (10 U/ μ l). Indicate the minimum total reaction volume required for digesting 10 μ g of plasmid DNA and the order in which these reagents should be added. (4 marks)
- (c) Explain briefly the effect of inserting a 1.0 kb random gene fragment into EcoRI polycloning site in pUC19 on the plasmid function. (7 marks)



(7 marks)

- (d) Calculate the expected sizes of the DNA fragments from the following digests of pUC19:
- (i) BamH1
- (ii) Sca1 and PvuII

(3 marks)

...6/-

1. Jawab semua bahagian soalan ini.

(a) Jelaskan fungsi “promoter” di dalam vektor bakteria.

(2 markah)

(b) Sekumpulan gen di bawah kawalan satu “promoter” digelar “operon”. Jelaskan “lac operon” dengan bantuan gambarajah.

(4 markah)

(c) Jelaskan dan lakarkan fungsi mekanisma kawalan penyah-rangsang laktosa apabila: -

(i) Laktosa hadir

(7 markah)

(ii) Laktosa tidak hadir

(7 markah)

2. Jawab semua bahagian soalan ini.

(a) Terdapat tiga jenis penyediaan DNA. Namakan dan jelaskan dengan ringkas ketiga-tiga jenis persediaan DNA ini.

(12 markah)

(b) Terangkan proses penulenan DNA menggunakan kromatografi penukaran ion.

(8 markah)

3. Jawab semua bahagian soalan ini.

(a) Terangkan tiga tahap utama dalam dogma biologi molekular.

(3 markah)

(b) Terangkan fungsi molekul RNA berikut.

(i) mRNA

(ii) tRNA

(iii) rRNA

(3 markah)

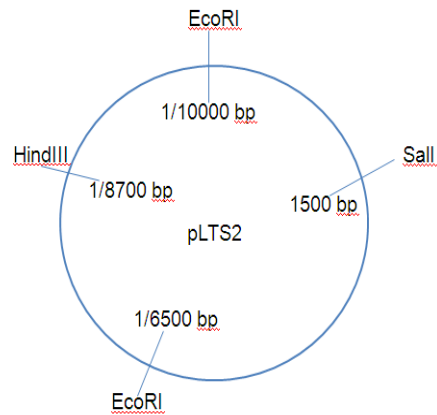
(c) Urutan DNA yang diberikan adalah seperti di bawah:
5'-TAC CCC ATT CAT TAC CCT ATC-3'

(i) Apakah urutan DNA komplementarinya?

(ii) Apakah urutan mRNAnya?

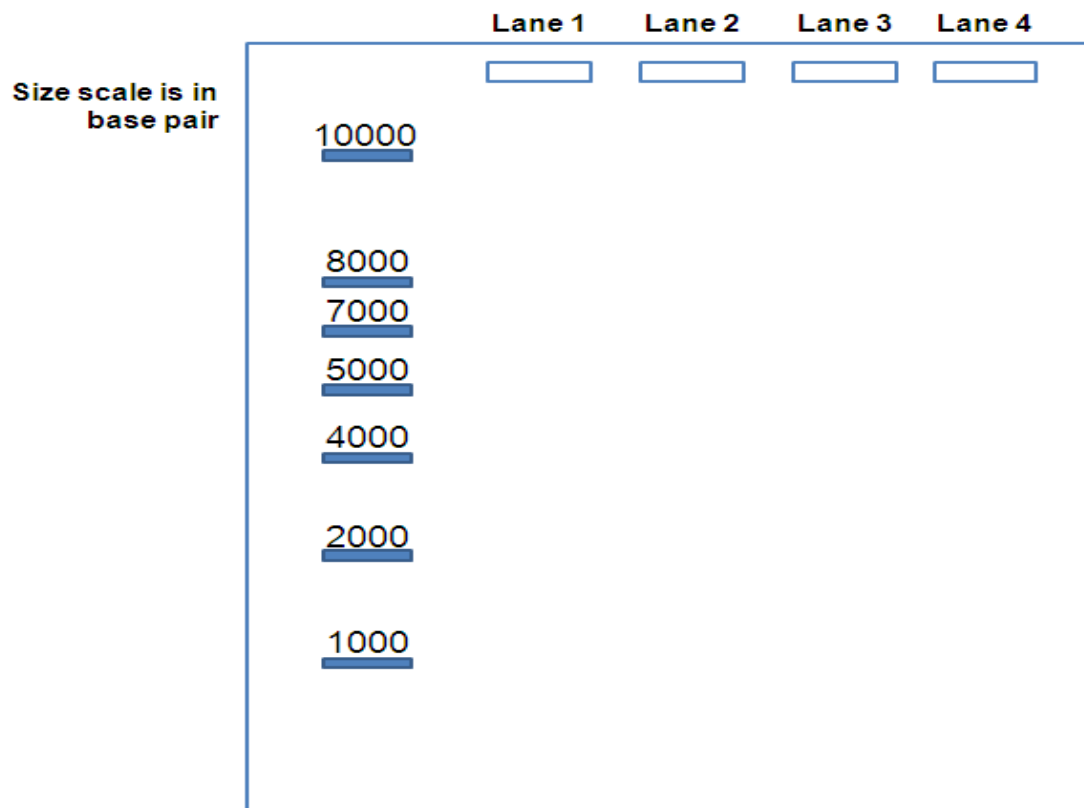
(2 markah)

- (d) Menggunakan peta vektor yang diberi, lengkapkan gel elektroforesis di bawah dengan potongan-potongan enzim berikut.



- (i) Lorong 1: potongan hanya dengan *EcoRI*
- (ii) Lorong 2: potongan dengan *Sall* dan *HindIII*
- (iii) Lorong 3: potongan dengan *Sall* dan *EcoRI*
- (iv) Lorong 4: potongan menggunakan kesemua enzim

(12 markah)



4. Jawab semua bahagian soalan ini.

(a) Jelaskan tiga tahap utama (denaturasi, pelekatan dan sambungan) dalam tindakbalas rantai polimerase.

(9 markah)

(b) Huraikan dua parameter penting untuk dipertimbangkan apabila menjalankan eksperimen PCR.

(6 markah)

(c) Kirakan bilangan molekul DNA yang hadir di dalam 1 ng plasmid dengan saiz 5 kb. Saiz purata nukleotida ialah 649 dalton.

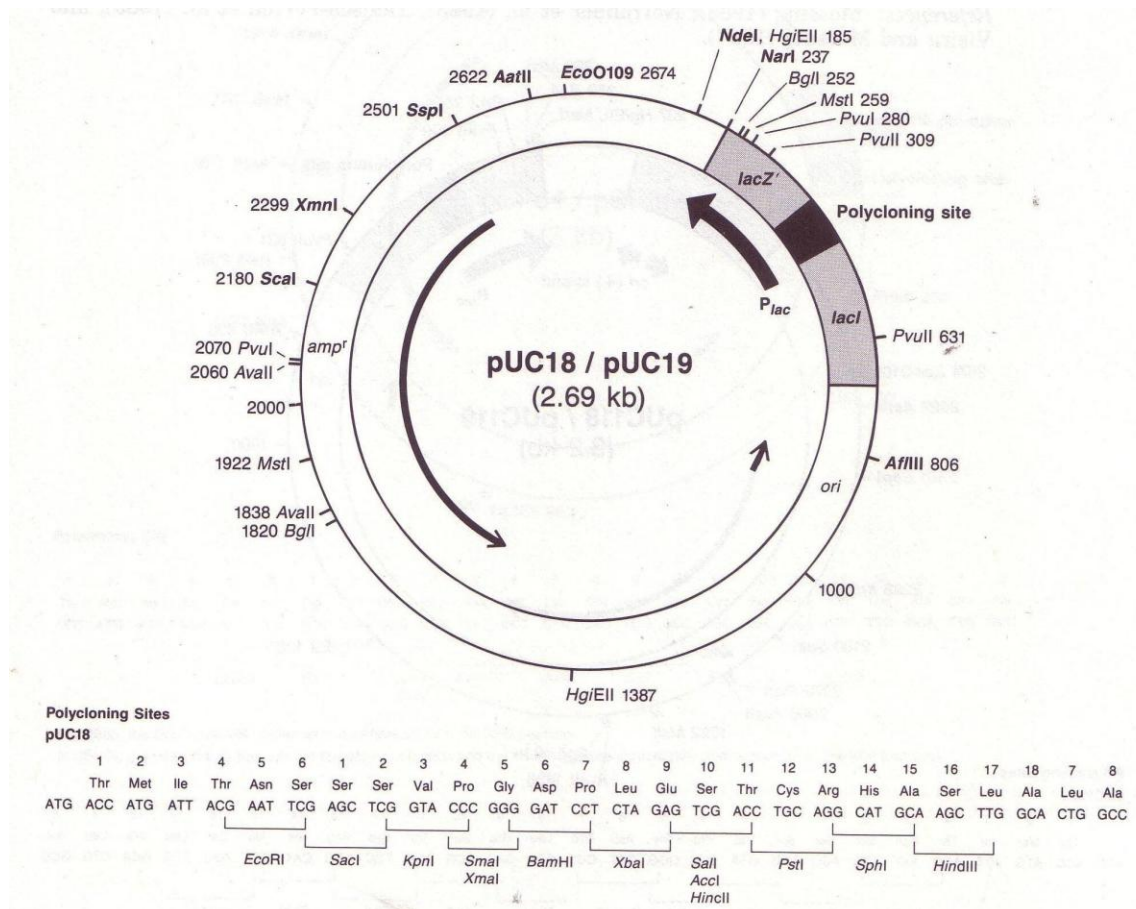
(5 markah)

5. Jawab semua bahagian soalan ini.

(a) Jelaskan kesignifikanan reagen berikut seperti yang digunakan untuk prosedur pengekstrakan DNA total bakteria: lizozim, RNase dan etanol mutlak.

(6 markah)

- (b) Sediakan campuran tindakbalas cerna sekatan yang mengandungi komponen daripada larutan stok berikut: DNA plasmid (2 mg/ml), penampun tindakbalas (10x), dan BamHI (10 U/μl). Nyatakan isipadu tindakbalas total minimum yang diperlukan untuk mencerna 10 μg DNA plasmid dan turutan reagen ini patut ditambah. (4 markah)
- (c) Terangkan secara ringkas kesan memasukkan fragmen rambang gen 1.0 kb ke dalam tapak polipengklonan EcoRI dalam pUC19 ke atas fungsi plasmid. (7 markah)



(7 markah)

- (d) Kirakan saiz fragmen DNA yang dijangkakan daripada cernaan pUC19 berikut:
- (i) BamHI
- (ii) SacI dan PvuII

(3 markah)