THAPAR INSTITUTE OF ENGINEERING AND TECHNOLOGY: PATIALA

(Department of Biotechnology & Environmental Sciences)

End-Sem Examination (Sem-I, 2006-2007), B.Tech. Biotechnology 4th Year

Genetic & Metabolic Engineering (BT-010)

Maximum Marks: 36 **Time: 3.0 Hrs**

09-12-2006

Attempt all the questions.

(Answers should be precise and to the point. Provide diagram if required to improve the quality of the answers.)

1. Attempt ***any three*** of the following questions: **2.0x3**

   a) A typical ligation reaction leads to varying products. Justify the statement.
   b) Calculate the amount of DNA present in 0.5 ml solution having A_{260}~0.4.
   c) You are given λ DNA digested with *Hind*III enzyme. How do you demonstrate the test ligation using this DNA sample?
   d) There is a mixture of two DNA fragments 'A' & 'B' of equal size i.e., 2.5 kb. The fragment 'B' contains an internal *Hind*III site. How do you purify the fragment 'A' from this mixture?

2. a) Give a diagram of plasmid-based *E. coli* expression vector. What are the factors that affect the expression of cloned genes in *E. coli*? **1.0+2.0**
   b) Give an example of inducible promoter that drives foreign genes in *E. coli*. Give your comments on its usefulness. **2.0**
   c) Only name any four protein products of recombinant DNA technology. **1.0**

   OR

   a) What are the ingredients required for Sanger's method of DNA sequencing? Also mention briefly the underlying principle. What is automated DNA sequencing **1.0+2.0 +1.0**
   b) What is the purpose of 'gel retardation assay'? How do you execute the same? **2.0**

3. a) What are the commonly used molecular techniques for gene expression studies? Outline the steps involved in any one of them. **2.0**
   b) Define 'gene library' and 'chromosome walking'. **2.0**
   c) Write a short note on *in vitro* packaging of bacteriophage λ. **2.0**

   OR

   a) What are the desirable properties of a cosmid vector? Give in detail the cloning strategy in a cosmid vector. **1.5+2.5**
   b) How do you check the quality of a gene library? **2.0**

See the overleaf

4. Write a short note along with application of the following *(any three)*: **2.0x3**

   a) *E.coli* DNA Polymerase I
   b) *Bam*HI Methylase
   c) T4 Polynucleotide Kinase
   d) T7 RNA Polymerase

5. a) Suppose you have a completely sequenced 1275 bp cDNA sequence. Sequence analysis reveals an ORF from the bases 117 to 746 in one of the strands. Calculate the size and approx. molecular weight of the polypeptide encoded by this predicted ORF. **2.0**
   b) What are the characteristic features of eukaryotic mRNA. How do you purify this mRNA population? **2.0**
   c) What are the stringency parameters commonly employed during nucleic acid hybridization? **2.0**

6. a) Sometimes BSA is added to restriction digestion reaction. Explain why? **1.5**
   b) How do you prepare 'DNAse-free RNase' solution? **1.5**
   c) How do you establish host restriction through bacteriophage plating assay? **1.5**
   d) What is the advantage of partial digestion of genomic DNA sample? **1.5**

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