Name : .....

Roll No. : .....

Invigilator's Signature : .....

CS/M.TECH(BT/PHMB/PHMC)/SEM-2/MBT/PHMB/PHMC-201/2012

2012

GENETIC ENGINEERING / RECOMBINANT DNA TECHNOLOGY

Time Allotted : 3 Hours *Full Marks* : 70

The figures in the margin indicate full marks.

Candidates are required to give their answers in their own words as far as practicable.

GROUP – A

(Objective Type Questions)

1. Answer the following questions : $10 \times 1 = 10$

   a) Which group of endonucleases (restriction enzymes) comprises two different enzymes for modification or cleavage yet recognize the same symmetrical target sequence ?

   b) Which viruses are used as high-efficiency vectors for long-term, stable gene expression ?

c) Choose the correct alternative :

A blood sample from an individual will produce a unique DNA fingerprint, also called a DNA profile. This is because each individual has a unique set of

i) proteins in his/her cells

ii) mutations in his/her DNA

iii) RNA in his/her nuclei

iv) enzymes in his/her mitochondria

v) amino acids in his/her blood.

d) Give examples of neoschizomers.

e) Name an appropriate means to terminate a growing DNA chain.

f) Ampicillin does not affect existing cells with intact cell envelopes but kills dividing cells as they synthesize new peptidoglycan. Write True or False.

g) Name a technique to study DNA-protein interactions.

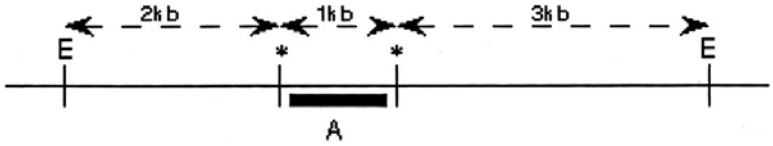
h) In Real time PCR, Sybr green method is more reliable in monitoring DNA synthesis/product formation in comparison to Molecular Beacon method.

Write True or False.

i) Name two compounds used as cryoprotectants.

j) Choose the correct alternative :

A particular RFLP is diagrammed below. *'E'* represents invariant EcoRI restriction sites. '*' represents polymorphic EcoRI sites. The dark box represents the location of a particular DNA probe *'A'*. What are all the possible alleles (i.e. size of bands) seen on a Southern blot probed with *'A'* ?



i) 1 kb, 2 kb, 3 kb, 4 kb, 5 kb, 6 kb

ii) 1 kb, 3 kb, 4 kb, 6 kb

iii) 3 kb, 4 kb, 6 kb

iv) 2 kb, 3 kb, 6 kb

v) None of these.

GROUP – B

Answer any *six* questions. $6 \times 10 = 60$

2. a) "Usually antisense RNA acts as positive regulatory molecule in regulating the copy number of relaxed plasmids." Explain the statement with special reference to ColE1-derived plasmids. 4

b) Explain very briefly what you understand by an ideal cloning vector. Why should an ideal cloning vector be small in size ? 3

c) How would you utilize selectable marker genes and markers for screening in DNA cloning experiments ? 3

3. a) Describe the factors affecting the decision for lysogeny or lytic infection cycle in lambda phase. How is this decision regulated ? 4

b) Explain any *three* of the following : 3 × 2

i) Why replacement vectors have higher cloning capacity in comparison to insertional vectors?

ii) Why Mutation in host *hfl* locus favours lysogeny ?

iii) Why are recombinants formed while cloning in replacement vectors *Spi* – ?

iv) What do you understand by incompatibility of plasmids ?

4. Write short notes on any *four* of the following : 4 × 2½

a) Pyrosequencing

b) Star Activity

c) YAC

d) Oligo capping

e) In vivo Excision

f) RNAase Protection Assay.

5. a) The presence of poly (*A*) tail in eukaryotic *m*RNAs facilitates isolation as well as *c*DNA synthesis. Elaborate on the statement. 4

   b) Why is it necessary to convert *m*RNA to *c*DNA for cloning into vector ? 1

   c) What are the advantages of RNA probes ? Give examples of a vector that can be used for the preparation of RNA probes. 5

6. a) You have isolated a gene for an enzyme that is expressed in *E. coli*. Describe how you would alter the activity of the enzyme. Assume that you know the DNA sequence of the gene but do not know anything about which regions of the gene are important for catalytic activity. 4

   b) Bisulphite mutagenesis is used for introducing localized point mutagenesis. To perform this you need to generate the single stranded regions that will be susceptible to bisulphite treatment. Describe the procedure with a technique that you know. 4

c) Regulating the activity of Ribonucleuotide Reductase may elevate the spontaneous mutation rates. Explain with an example. 2

7. a) What are reporter genes and how are they useful for promoter analysis ? Explain with a suitable example. 4

b) Explain schematically how you would introduce foreign DNA using embryonic stem cells. 4

c) Illustrate with a suitable example how a transgene expression can be targeted to specific tissues. 2

8. a) What is a microsatellite ? Give a very brief description. 3

b) What types of DNA methods are RAPD and RFLP ? Describe briefly how RAPDs differ from more standard applications of this type of method. 4 + 1

c) How can microarray be used for comparative studies ? 2

9. a) What is the principal mechanism of antisense RNA technology ? 2

b) By utilizing a selectable marker how would you successfully achieve gene silencing in mammalian cells ? 4

c) How would you utilize transgenic microorganisms for the production of recombinant therapeutic proteins or recombinant biopharmaceuticals ? 4

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