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Term End Examination 2025-2026
Programme – M.Sc.(BT)-2025
Course Name – Advances in Genetic Engineering
Course Code - MBT10402
(Semester I)

Full Marks : 60 **Time : 2:30 Hours**

[The figure in the margin indicates full marks. Candidates are required to give their answers in their own words as far as practicable.]

Group-A
(Multiple Choice Type Question) 1 x 15=15

1. *Choose the correct alternative from the following :*

(i) R-loop analysis is best suited for detecting:
a) RNA splicing
b) DNA methylation
c) Gene expression levels
d) Introns in a gene

(ii) In DNA footprinting assay, the region protected from cleavage indicates:
a) Unmethylated CpG island
b) Promoter region
c) Protein binding site
d) Open reading frame

(iii) Chromosome walking involves:
a) Overlapping clones to traverse genomic DNA
b) Mutant rescue via homologous recombination
c) In situ hybridization of genes
d) PCR with variable primers

(iv) Plasmids are used for carrying out the cloning procedure. Which of the statement is true for plasmids?
a) Bacterial plasmids are linear in nature
b) They are single stranded
c) Insertion of DNA into plasmid allows it to be propagated in host cells and they are known as vectors because of their this property
d) They are not capable of replication in bacteria

(v) Phagemid consist of
a) Plasmid vector carrying λ phage's cos site
b) plasmid and M13 components
c) Plasmid vector carrying origin of replication of λ phage only
d) Plasmid vector carrying origin of replication of plasmid only

(vi) Choose the formula to calculate Melting temperature of a DNA template
a) 4(G+C) + 2(A+T)
b) 2(G+C) + 4(A+T)
c) 2(A+G) + 4(C+T)
d) 4(A+G) + 2(C+T)

(vii) In order to clone an eukaryotic gene in pBR322 plasmid vector, the desired DNA fragment was produced by PstI cleavage and incubated with PstI digested pBR322 (PstI

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cleavage site lies within the ampicillin resistant gene) and ligated. Mixture of ligated cells were used to transform E. coli and plasmid containing bacteria were selected by their growth in tetracycline containing medium. Predict which type of plasmids will be found?

a) Circular pBR322 plasmid containing the target gene and resistant to only tetracycline (tet)

b) Circular pBR322 plasmid containing the target gene and resistant to only tetracycline (tet) only and recircularized pBR322 plasmid resistant to both ampicillin (amp) and tet

c) Circular pBR322 plasmid containing the target gene and resistant to only tetracycline (tet) only, recircularized pBR322 plasmid resistant to both ampicillin (amp) and tet and concatemerized pBR322 resistant to both amp and tet

d) Circular pBR322 plasmid containing the target gene and resistant to both amp and tet

(viii) The order for the construction of a cDNA fragment from mRNA is to

a) bind oligo-dT, treat with reverse transcriptase, digest with RNase, add G residues to the 3' end, bind oligo-dC, treat with DNA polymerase

b) treat with reverse transcriptase, digest with RNase, add G residues to the 3' end, bind oligo-dC, treat with DNA polymerase and bind oligo-dT

c) digest with RNase, add G residues to the 3' end, treat with reverse transcriptase, add G residues to the 3' end and treat with DNA polymerase

d) bind oligo-dC, treat with reverse transcriptase, digest with RNase, add G residues to the 3' end, bind oligo-dT and treat with DNA polymerase

(ix) Taq polymerase is a _____ polymerase.

a) heat-stable
b) buffering
c) denaturant
d) large

(x) In blue-white selection of recombinant bacteria, the transformed colony with the insert will be_____. Select from the given options

a) colourless
b) black
c) blue
d) white

(xi) Divalent cations are used in PCR reaction mixture:

a) as a co-factor
b) as an inhibitor
c) as a catalyst
d) none of the these

(xii) Reverse Transcription PCR is used to

a) RNA strand is reverse transcribed into its complementary DNA
b) amplify DNA sequence
c) quantify the copy number of nucleotide sequences
d) All of these

(xiii) Ligase joins the nicks by creating

a) Covalent bond
b) Phosphodiester bond
c) Hydrogen bond
d) Ionic bond

(xiv) One of the most significant discoveries which allowed the development of recombinant DNA technology was:

a) the discovery of antibiotics used for selecting transformed bacteria

b) the identification and isolation of restriction endonucleases permitting specific DNA cutting

c) the discovery of DNA and RNA polymerase allowing workers to synthesize any DNA sequence

d) the development of the polymerase chain reaction

(xv) Choose from the following that must be true for a nucleic acid probe to hybridize

a) The sequence to which it hybridizes must be identical to that of the probe.

b) The sequence which it hybridizes must be complementary to the probe

c) Both the probe and the sequence to which it hybridizes must be DNA

d) Neither the probe nor the sequence to which it hybridizes can contain introns

Group-B

(Short Answer Type Questions) 3 x 5=15

2. We use chilled ethanol and 1/10th volume of Sodium acetate (pH 5.2) for DNA precipitation. Discuss the principle behind this phenomenon (3)
3. 1% polyvinylpyrrolidone is one of the important reagents in extraction buffer during plant genomic DNA isolation, however in case of E.coli it is not required. Explain why (3)
4. A researcher has only 5'-end labeled DNA and wants to determine its sequence without using DNA polymerase. Which sequencing method should they use and why? (3)
5. Analyze how the selection of restriction enzymes impacts the success of directional cloning. (3)
6. Design a strategy to optimize a eukaryotic gene for expression in a prokaryotic host. What elements would need modification? (3)

OR

Evaluate the importance of promoter and terminator sequences in the design of an efficient expression vector. (3)

Group-C

(Long Answer Type Questions) 5 x 6=30

7. Analyze why TaqMan probe-based qPCR assays are preferred over SYBR Green dye-based assays in the development of diagnostic kits, particularly with respect to specificity, multiplexing, and clinical reliability. (5)
8. Explain the function of Solution I, II and III in plasmid isolation procedure (5)
9. Explain how Sanger sequencing can be adapted for high-throughput analysis of a microbial community, and discuss why Maxam-Gilbert is unsuitable for such studies. (5)
10. A gene encoding for a novel protein is cloned in pBR322 and transformed in E. coli strain DH5α. Develop rDNA process for this objective and explain how do you select positive recombinants? (5)
11. List the advantages and disadvantages of using plasmids and YACs as cloning vectors (5)
12. Differentiate between Cosmids and phagemids (5)

OR

Analyze the roles of detergents, chloroform, sodium salts, EDTA and isopropyl alcohol in nucleic acid isolations (5)

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