Register Number:

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ST. JOSEPH'S COLLEGE (AUTONOMOUS), BENGALURU-27
M.Sc. MICROBIOLOGY- III SEMESTER
SEMESTER EXAMINATION -NOVEMBER 2020
MB 9118 – RECOMBINANT DNA TECHNOLOGY

Time: 2 1/2 hrs **Max. Marks - 70**

This paper contains **2** printed pages and **4** parts

I. Answer any Five of the following **5x3=15**

1. List the steps used to prepare a recombinant DNA molecule.
2. What is cDNA? Why is it necessary to generate cDNA before cloning and expressing a eukaryotic gene in a bacterium?
3. Differentiate selection versus screening of recombinants with suitable examples.
4. What are mini cells? List its application.
5. What impact does the release of recombinant DNA has on environment?
6. Contrast Somatic cell gene therapy with germline gene therapy.
7. What is chromosome walking? List its applications.

II. Answer any Five of the following **5x5=25**

8. Explain how restriction enzymes recognize and digest DNA tocreate either blunt or sticky ends. Give suitable examples.
9. Given pUC8 plasmid as a vector, design a gene cloning experiment using the same and explain how recombinants are selected?
10. What signalling elements in a vector help express a cloned gene in a bacterial host? Describe affinity chromatography used for purification of expressed proteins from bacteria.
11. Describe the principle and methodology involved in electroporation.

12. What is the purpose of Southern blotting? How is a probe selected? Why do you think the Southern blotting technique wasan important breakthrough when it was introduced?

13. Illustrate Genomic library construction. What are the applications of Genomic Libraries?

14. What are transgenic plants? How are insecticide resistant plants generated?

III. Answer any Two of the following 2X10=20

15. a. Explain why PCR results in the amplification of a specific DNA sequence despite many competing sequences.3
    b.Contrast Multiplex PCR with Gradient PCR. 4
    c. Summarize the importance of PCR in biology3

16. a. Explain how DNA is sequenced by the Sanger Coulson's terminationmethod.5
    b. List the types of microarrays. Write their applications 5

17. a.Illustrate chemical synthesis of DNA5
    b. How are bacterial cells made competent by artificial means in calcium chloride mediated gene transfer? 5

IV. Answer the following 1X10=10

18. a. You are studying chemotaxis proteins in a newly describedbacterium. You have cloned a gene that encodes the CheA protein. You need to be sure that this protein localizes to the inside of the plasma membrane. Will you use a transcriptionalor translational fusion? Explain your choice. 3

    b. You want to visualize a digested plasmid that yields fragments of 100 bp, 400 bp, and 3,000 bp. You have another plasmid digested with the same endonuclease that yields two fragments, 4,000 bp and 5,500 bp. How many recognition sites are there for this enzyme in each plasmid? For which plasmid would you use a 1.5% agarose gel? For which would a 0.8% gel be best? Explain your answer. 3

    c. Calculate the Tm for the FWD and RVS primers given below and suggest an ideal annealing temperature if these primers are used subsequently. 4

    Forward Primer (FWD) - 5' gcgcatatatcgcgtatatagag 3'

    Reverse Primer (RVS) - 5' gagatatatgcgctatatacgcg 3'