MANIPAL INSTITUTE OF TECHNOLOGY
MANIPAL
(A constituent unit of MAHE, Manipal)

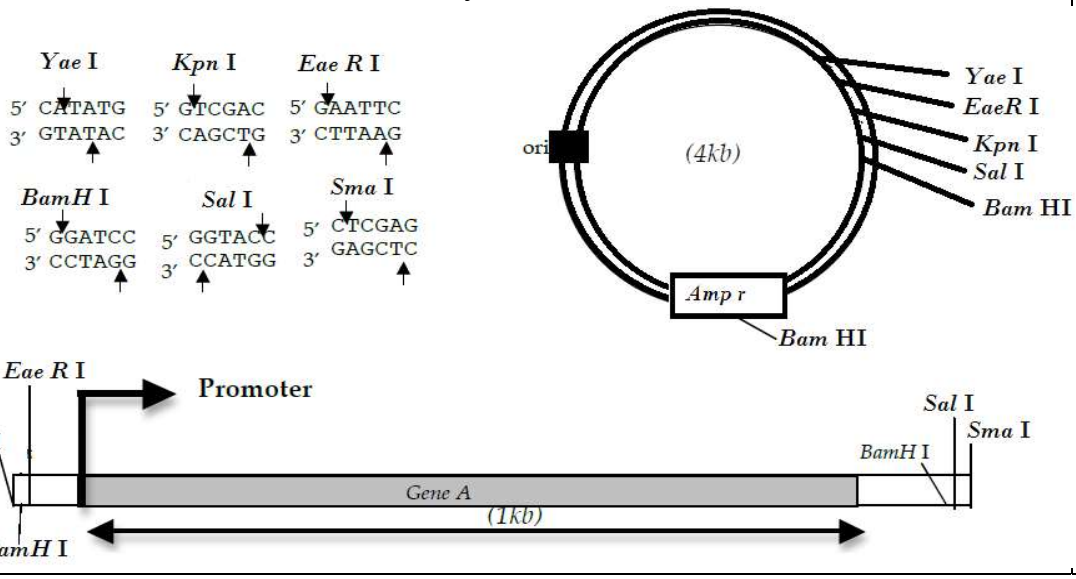
IV SEMESTER B.TECH. EXTERNAL EXAMINATIONS APRIL 2019

SUBJECT: GENETIC ENGINEERING [BIO 2203]

Date of Exam: **26/04/2019** Time of Exam: **2.00 PM – 5.00 PM** Max. Marks: **50**

Instructions to Candidates:

- Answer ALL the questions & missing data may be suitable assumed

1A.	In what ways are human artificial chromosomes (HACs) better than viral based vectors? Is it correct to say that synthetic biologists may use HACs to hold entire man-made biological pathways? Explain.	**5**
1B.	Elaborate on cosmids.	**5**
2A.	The recognition sites for several restriction enzymes are shown below. *gl*I 5GA/TC3 3CT/AG5 *Pst*I 5A/GATCT3 3TCTAG/A5 *Bcl*HI 5G/GATCC3 3CCTAG/G5 *Bam*I 5T/GATCA3 3ACTAG/T5 Consider the following small DNA sequence 5′–CGCACCTGTGTTGATCACCTAGCCGATCCACGGTGGATCCAAGGC–3′ 3′–GCGTGGACACAACTAGTGGATCGGCTAGGTGCCACCTAGGTTCCG–5′ For each of the restriction enzymes listed, give the number of times that the enzyme will cut the DNA fragment above. Also give the number of resulting DNA fragments after individual treatment with each enzyme.	**2**
2B.	How does DNA *Ligase* work in joining two heterogeneous DNA fragments?	**3**
2C.	Expand TdT, Explain the TdT mechanism of action	**3**
2D.	The plasmid vector, Gene A and their restriction sites are given below. Construct a recombinant DNA (plasmid + Gene A) using the right set of restriction enzymes. List the combination of enzymes. 	**2**

<table>
<tr><td>3A.</td><td>What could be the reasons for weak signal in fluorescence in situ hybridization experiments</td><td>4</td></tr>
<tr><td>3B.</td><td>What is the significance of prehybridization step in southern blotting</td><td>3</td></tr>
<tr><td>3C.</td><td>A smaller amount or decrease in time of DNA exposure to restriction enzyme was followed while constructing a gDNA library. Explain why this approach may allow you to construct a more complete library?</td><td>3</td></tr>
<tr><td>4A.</td><td>How does the chemical cleavage method work for sequencing DNA molecule?</td><td>3</td></tr>
<tr><td>4B.</td><td>Explain the importance of linker in GE with illustration.</td><td>2</td></tr>
<tr><td>4C.</td><td>Which parameter would you change first, if your PCR reaction gives too many products? What would you do if the PCR reaction gives very little, if any, of the correct product?</td><td>3</td></tr>
<tr><td>4D.</td><td>The λ bacteriophage genome (double stranded, linear DNA, 48,500 bp size) labeled at 5’ end with a radioactive phosphorous () was digested with different restriction enzymes under conditions that permits partial digestion of the DNA. The resulting fragments were separated using agarose electrophoresis and bands were visualized by autoradiography. With the results shown in the below table, construct the λ phage restriction map. (2)
<table>
<tr><th>Fragments</th><th>Apa I (bp)</th><th>Pvu I (bp)</th><th>Bam HI (bp)</th></tr>
<tr><td>1</td><td>48,500</td><td>48,500</td><td>48,500</td></tr>
<tr><td>2</td><td>10085</td><td>35,790</td><td>41,730</td></tr>
<tr><td>3</td><td></td><td>26250</td><td>34500</td></tr>
<tr><td>4</td><td></td><td>11,930</td><td>27,920</td></tr>
<tr><td>5</td><td></td><td></td><td>22,345</td></tr>
<tr><td>6</td><td></td><td></td><td>  5,505</td></tr>
</table></td><td>2</td></tr>
<tr><td>5A.</td><td>5-HT2A is a neuroreceptor, The following SNPs in 5HTR2A: Rs6313, Rs6311 and Rs6314 were found during clinical investigations. Discuss the 3 SNPs with respect to its change in nucleotide, synonymous / non-synonymous condition and its associated disease.</td><td>3</td></tr>
<tr><td>5B.</td><td>Brain tumor (BT) is abnormal growth of cells within the brain. Exposure to vinyl chloride or ionizing radiation will mutate and delete tumor suppressor genes are the lead cause for BT. Temozolomidetype is an effective drug for the treatment of BT. Besides target specificity, toxicity is the major problem with the drug.
• Suggest an alternate method by which you can solve the above problem. (1)
• How different is your method from the conventional cancer therapies? (1)
• What do you need to know about the target disease in order to apply your method? (1)
• What do you think are some of the challenges facing your method? (1)</td><td>4</td></tr>
<tr><td>5C.</td><td>Ti Plasmid produces tumor in plants. Explain the role of genes present in virulence region for creating transgenic plant.</td><td>3</td></tr>
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