

U.G. 4th Semester Examination - 2021**Molecular Biology****[PROGRAMME]****Course Code : MBG-401-T-CC-10****(Recombinant DNA Technology)**

Full Marks : 20

Time : 1 Hour

*The figures in the right-hand margin indicate marks.**Candidates are required to give their answers in their own words as far as practicable.*

1. Answer any **five** questions: $1 \times 5 = 5$
- a) What is the role of calcium chloride in preparation of *E. coli* competent cell?
 - b) What is Chromosome walking?
 - c) State few applications of knockout mice in biotechnology research?
 - d) What is a Cosmid vector?
 - e) Why selection markers are so important during the process of cloning?
 - f) Why do we give heat shock for uptake of foreign DNA by host competent cell?
 - g) What are oligonucleotide drugs?

- h) What is the application of RDT in pharmaceutical industry?

2. Answer any **one** of the following: $5 \times 1 = 5$
- a) Why does a linear plasmid migrates more in an Agarose gel than a nicked plasmid? Even if the entire Agarose gel contains Ethidium bromide, only the DNA band fluoresces under UV transilluminator. Why? $2\frac{1}{2} + 2\frac{1}{2}$
 - b) In *E. coli* BL21 DE3 expression host, T7 RNA polymerase is under Lac promoter and in the suitable plasmid vector, gene of interest is under T7 promoter for induction by IPTG. Discuss the advantages of this system and the consequences of directly placing the gene of interest under Lac promoter?
 - c) Differentiate between a genomic DNA and a cDNA library. How do we retrieve clones from DNA libraries? $3 + 2$
3. Answer any **one** of the following: $10 \times 1 = 10$
- a) Using different blotting techniques, how can you determine the loss of function mutations (tumour suppressor gene). Under what conditions a Restriction endonuclease exhibit star activity? How many fragments can you

expect after digestion of a 50,000 bp DNA by a 6(bp) cutter restriction endonuclease?

6+2+2

- b) How can you quantitatively determine the amplification of DNA in real time using PCR? If the forward and reverse primers have very high or low annealing temperatures or have complimentary regions among themselves, what are the problems that you might face during the PCR? What kind of experiment would enable you to determine the binding region of a transcriptional regulator within a DNA? How can you insert a point mutation in middle of a gene using PCR and primers? 2+2+3+3
- c) Retroviruses and adenoviruses are preferred vectors for gene therapy but may have severe adverse effects. Explain. In spite of so many advancements, why the success rate of gene therapy is still very low? How DNA fingerprinting can be used to identify criminal or solve paternity disputes? Can you use DNA fingerprinting when the crime suspects are identical twins? 2+3+4+1
