2054

M.E. (Bio-Technology) Second Semester ME-BIO-204: Genetic Engineering

Time allowed: 3 Hours

Max. Marks: 50

NOTE: Attempt <u>five</u> questions in all, including Question No. I which is compulsory and selecting two questions from each Unit.

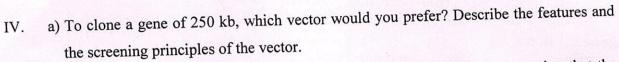
x-x-x

- I. Attempt the following:
 - a) In gene-cloning experiments, why is the cleaved plasmid DNA often treated with alkaline phosphatase prior to the ligation step?
 - b) What enzymatic activities are associated with Klenow fragment?
 - c) What are degenerate primers?
 - d) What is touchdown PCR?
 - e) What are transposable elements?
 - f) What are linked genes?
 - g) What is the major limitation of using retroviral vectors to deliver genes into the cells?
 - h) What is a negative selection marker? Give an example.
 - i) What is a bacmid?
 - j) The heterologous expression of a restriction enzyme in *E. coli* results in degradation of the host DNA. Suggest a way to circumvent this problem. (10x1)

UNIT-I

- II. a) Describe the potential problems in expressing a gene from a eukaryote in E. coli.
 - b) Differentiate between genomic library and cDNA library (atleast 5 points). (2x5)
- III. a) Differentiate between class I and class II restriction enzymes (atleast 4 points).
 - b) Describe the general features of plasmid vectors.
 - c) What is inverse PCR? What is it used for?

(4,4,2)



b) A gene for an enzyme has been isolated and expressed in *E. coli*. Assuming that the DNA sequence of the gene is known but nothing is known about the residues that are important for catalytic activity, describe how the catalytic activity of the enzyme can be altered. (2x5)

UNIT - II

- V. a) Describe and discuss the PCR/OLA detection protocol for genetic diseases.
 - b) Describe Agrobacterium tumefaciens mediated gene transfer in plants. (2x5)
- VI. a) What is a molecular beacon probe? How does it work? How can molecular beacon probes be used to detect several genes in the same sample?
 - b) What are humanized monoclonal antibodies? Describe how they are generated. (2x5)
- VII. a) Explain how RNA interference is used for improving disease resistance in plants.
 - b) Describe the methodology of RAPD for Marker Assisted Selection. Outline the advantages, limitations and applications of RAPD. (2x5)