JAYPEE UNIVERSITY OF INFORMATION TECHNOLOGY, WAKNAGHAT

TEST -3 EXAMINATION- JUNE 2023

M.Sc. (Microbiology) II Semester

COURSE CODE (CREDITS): 18MS1BT313 (3) MAX. MARKS: 35

COURSE NAME: RECOMBINANT DNA TECHNOLOGY

COURSE INSTRUCTORS: Dr. Rahul Shrivastava MAX. TIME: 2 Hours

Note: (a) *All questions are compulsory.*

(b)Marks are indicated against each question in square brackets.

Q1. GDP mannose gene of *Pseudomonas aeruginosa* needs to be amplified which produces a specific 520-bp PCR product useful for *P. aeruginosa* identification. Primers used for the amplification are:

Forward Primer - 5' TACCGTCAGCGGCACATAGAT

Reverse Primer - 5' TAGAACATGACTGCCCGGTTCA

a. Calculate the Tm of the primers. **[2]**

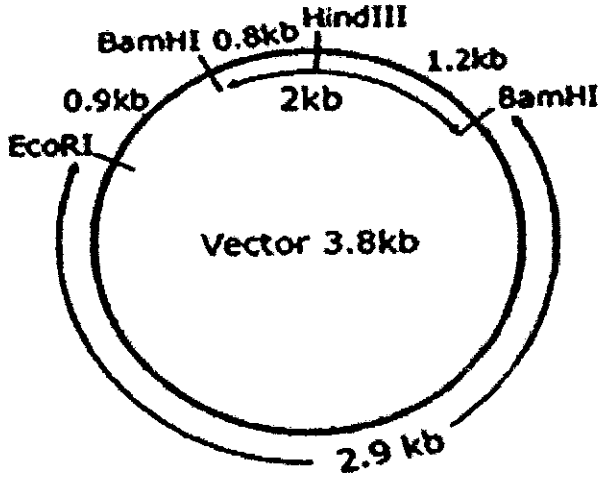
b. Design a PCR cycle for amplification of the 520bp product **[3]**

c. Calculate the amount of PCR product that would be obtained after 5 cycles if the initial amount of gene product is 25ng. **[2]**

Q2. Gene expression studies need to carried out for *Homo sapiens* and *Escherichia coli*, which library type would be construct for each species containing the gene pool, for expression analysis. Design the protocol for each library type, with detailed steps. Compare the advantages and limitations of each library type. **[7]**

Q3. Amylases enzyme is produced for commercial use using recombinant DNA technology. Describe in detail the use of **'histidine'** and **'Mbp (maltose binding protein)'** tags for purification of the protein. Compare the advantages and disadvantages of the ***his*** and ***mbp*** tags for such purification. **[7]**

Q4. The diagram represents a 2 Kb insert successfully introduced between two BamHI sites of a 3.8 Kb vector in the desired orientation. The HindIII sites on the insert and EcoRI site on the vector are also indicated.



a. Size of DNA product obtained after digestion with EcoRI [1]
b. Size of DNA product obtained after digestion with BamHI [1]
c. Size of DNA product obtained after digestion with HindIII [1]
d. Size of DNA product obtained after double digestion with EcoRI and HindIII [1]
e. Draw a well labeled agarose gel showing different product/digested product(s) obtained from a, b, c, and d. [3]

Q5. Write Short Notes on the following:

a. *Agrobacterium rhizogenes* mediated gene transfer [2]
b. *Agrobacterium tumefaciens* mediated gene transfer [2]
c. *Bt-Cotton* [3]