



B.TECH DEGREE EXAMINATIONS: NOV/DEC 2022

(Regulation 2018)

Fifth Semester

BIOTECHNOLOGY

U18BTI5201: Genetic Engineering and Genomics

COURSE OUTCOMES

- CO1:** Comprehend and choose cloning steps for recombinant DNA construction
CO2: Analyze the features of various types of gene cloning vectors and design a suitable vector for recombinant protein expression
CO3: Interpret various types of gene isolation and screening methods
CO4: Apply suitable modern molecular techniques to solve real life problems
CO5: Evaluate regulatory issues of GMOs and their environmental and societal impact
CO6: Analyze and interpret various genome analysis methods

Time: Three Hours

Maximum Marks: 100

Answer all the Questions: -

PART A (10 x 1 = 10 Marks)

1. Match the type of DNA modifying enzyme (list I) and their function (List II)

CO1 [K₁]

List I	List II
A. Alkaline phosphatase	i. addition of nucleotides to the 3' end of the single stranded DNA
B. Polynucleotide kinase	ii. removing phosphate group
C. Terminal transferase	iii. addition of phosphate group
D. DNA ligase	iv. joining two DNA fragments

- | | A | B | C | D |
|----|-----|-----|-----|----|
| a) | ii | i | iii | iv |
| b) | iii | iv | ii | i |
| c) | ii | iv | iii | i |
| d) | ii | iii | i | iv |

2. In a blue-White screening method of recombinant selection, what do blue colonies represent?

CO2 [K₃]

- | | |
|-------------------------------|--------------------------------------|
| a) Cells NOT taken up the DNA | b) Non-Recombinant Clones |
| c) Recombinant clones | d) Cells with fragmented plasmid DNA |

3. Below are the salient features of a probe for Southern hybridization experiment. Choose the correct statement

CO4 [K₂]

- (s).
- Probe is a single stranded DNA or RNA
 - Probe is labeled doubled stranded DNA

PART B (10 x 2 = 20 Marks)
(Answer not more than 40 words)

11. Outline the step involved in synthesis of recombinant DNA. CO1 [K2]
12. Below is the typical structure of a eukaryotic gene. Identify the essential genetic elements shown as A, B, C and D. CO1 [K2]



13. Draw and label a cloning vector. CO2 [K2]
14. Provide all essential ingredients of typical Polymerase Chain Reaction (PCR) and their role. CO4 [K3]
15. Self-ligation of the vector DNA is a major problem in gene cloning. How will you overcome this challenge with suitable DNA manipulating strategy? CO2 [K4]
16. Calculate the number of clones to be screened from a genomic library with 98 % probabilities from the data given below. CO3 [K3]
- Size of the genome: 3×10^6 kbp
 Size of the DNA fragment: 25 kbp
 If you want to reduce the number of clones to be screened, which cloning vector you would suggest?
17. What are the goals and achievements of human genome project? CO6 [K2]
18. What is “C Value” paradox? Give any ONE example. CO6 [K2]
19. How is physical mapping of genome helped in successful completion of genome sequencing project? CO6 [K3]
20. DDRT PCR was performed with control and cancer cells. Below is the result of DD RT PCR gel image. Find out the difference in gene expression pattern. Suggest a suitable target for potential drug development. CO6 [K5]

Gene No.	Control Cell type	Cancer cell type
Gene1	_____	████████
Gene2	_____	_____
Gene3	_____	████████
Gene4	_____	_____
Gene5	_____	_____

Answer any FIVE Questions:-
PART C (5 x 14 = 70 Marks)
(Answer not more than 350 words)

21. a) Elaborate on construction of cDNA library from liver tissue to isolate liver specific genes and provide illustration on immunochemical screening method of cDNA library. 7 CO3 [K3]
- b) Justify why plasmid map is important in deciding the restriction sites for cloning a foreign gene. 7 CO2 [K4]

Construct a linear restriction map from the following data.

22 kbp DNA was restricted with KpnI, HindIII enzymes (single digest and double digestion)

KpnI: 11kbp, 6kbp, 5kbp

HindIII: 14kbp, 8 kbp

KpnI+HindIII: 8kbp, 6 kbp, 5kbp, 3 kbp

22. a) A foreign DNA of size 5.0kbp was cloned in lacZ part of the plasmid vector and transformed into bacterial (*E.coli*), upon selecting on IPTG+X-Gal+Amp selection plate, blue-white colonies were observed. The scientist picked out white colonies and started isolating recombinant DNA to confirm the presence of the insert. However, electrophoresis gel result showed that the plasmid does not contain foreign DNA. Analyze the situation and provide the suitable reason(s) 7 CO1 [K4]
- b) How is baculoviral vector used to express foreign gene in insect cell line? Add suitable illustration of AcMPV vector. CO2 [K2]
23. a) What is a “contig” in genome assembly. Explain any ONE method to close “Gaps” in the assembly. 7 CO6 [K2]
- b) “India has embarked on establishing various levels of biosafety laboratories and rDNA research guidelines” Evaluate this statement with respect to status of India with relevant points. 7 CO5 [K2]
24. a) How is CRISPR-Cas9 technology helpful to selectively remove or downregulate a particular gene expression? Can it be applied to human genome editing? 7 CO4 [K2]
- b) Compare “bottom up “and Top down methods of genome sequencing. Which method you would suggest to sequence “an elephant’s genome”? Why? 7 CO6 [K4]
25. a) Elaborate of *in situ* fabrication of microarray using photolithography and provide the steps involved in analysis of gene expression in control and cancer tissue samples using microarray experiment. 7 CO6 [K3]
- b) RNA interference (RNAi) is a technique used to silence a eukaryotic gene. Examine how RNAi can be performed to inactivate the gene of interest to get desired result. 7 CO4 [K3]
26. a) Explain a situation where Q-PCR can be used as molecular diagnostic tool for detection of diseases. 7 CO4 [K2]
- b) How are herbicide resistant transgenic plant developed? Whether herbicide resistant transgenic plant really helpful to Indian agriculture? Substantiate your answer. 7 CO4 [K3]
