P. P. SAVANI UNIVERSITY

Fifth Semester of B.Sc. Examination
December-2021
SSBT3150–Recombinant DNA Technology-II
13.12.2021, Monday Time: 12:30 p.m. to 03:00 p.m. Maximum Marks: 60

Instructions:
1. The question paper comprises of two sections.
2. Section I and II must be attempted in separate answer sheets.
3. Make suitable assumptions and draw neat figures wherever required.
4. Use of scientific calculator is allowed.

Section-I (Total Marks - 30)

Q.1 Short Questions **[10]**

1.1 Objectives **[05]**

1.1a cDNA stands for
- **A** Complex DNA
- **B** Complementary DNA
- **C** Corona DNA
- **D** All of the above

1.1b PACs vectors are based on:
- **A** Polyoma virus
- **B** Polio virus
- **C** P1 bacteriophage
- **D** Polyoma virus

1.1c Which of the following is/are applications of cDNA library
- **A** To study of gene function in-vitro
- **B** To determine alternative splicing in cells/tissues
- **C** Discovery of novel genes
- **D** All of the above

1.1d Digoxigenin is used for labeling of:
- **A** Proteins
- **B** DNA
- **C** RNA
- **D** All of the above

1.1e Which is true for Fosmids
- **A** it contains λ cos site
- **B** it contains F-plasmid origin of replication
- **C** it is a low copy number plasmid
- **D** All the options are true

1.1f During RNA extraction (at phenol cholofom step), at acidic PH i.e. PH 4.0
- **A** RNA will stay in aqueous phase and DNA will be in organic phase
- **B** RNA will stay in organic phase and DNA will be in aqueous phase
- **C** RNA will stay in the interphase and DNA will be in aqueous phase
- **D** None of the options are correct

1.1g Which of the following is potential challenge for genomic DNA library

construction

A Desired gene might be cut internally
B Gene size larger than vector capacity
C vectors have limited capacity to accommodate large insert DNA
D All of the options are true

1.1h BAC is

A a protein
B an enzyme
C a vector
D None of the above

1.1i Which of the following enzyme is used in end-filling reaction is

A Klenow fragment
B RNA Pol I
C Helicase
D Reverse Transcriptase

1.1j In blue white screening, WHITE colonies represent

A Transformed but non-recombinant cells
B Transformed and recombinant cells
C Untransformed and recombinant
D Untransformed and non-recombinant

1.2 Answer the Following: (MCQ/Short Question/Fill in the Blanks) **[05]**

1.2a What does UTR stands for?

1.2b M-MLV is a RNA dependent DNA polymerase (TRUE/FALSE)

1.2c During cDNA preparation genomic DNA contamination can be checked either by DNase or intron-exon crossing primers (TRUE/FALSE)

1.2d _____binds to streptavidin and avidin with an extremely high affinity and high specificity

1.2e What are Riboprobes?

Q.2 Short Notes (Attempt any two) **[06]**

A Manniatis strategy
B mRNA purification
C Blue white screening

Q.3 Explain in detail (Attempt any two) **[14]**

A What are the various steps of cDNA library? How cDNA library is different from Genomic library?
B Discuss three λ bacteriophage-based vectors in detail?
C What is a genomic library? Discuss its construction in detail.

Section-II (Total Marks - 30)

Q.1 Short Questions [10]

1.1 Objectives [05]

1.1a _____measures the direct release of H+ (protons) from the incorporation of individual bases

- **A** Illumina sequencing
- **B** Ion Torrent sequencing
- **C** Maxam- Gillbert sequencing
- **D** Solexa sequencing

1.1b In _____ each base emits a unique fluorescent signal, which is being recorded

- **A** Ion Torrent sequencing
- **B** Sanger sequencing
- **C** Illumina sequencing
- **D** Maxam- Gillbert sequencing

1.1c In Maxam-Gilbert sequencing method, pyrimidines are hydrolyzed using:

- **A** Dimethylsulphate
- **B** Hydrazine
- **C** Formic acid
- **D** Hot piperidine

1.1d What is the current prevalent method for genome sequencing projects

- **A** Chain Termination Method
- **B** Next-Generation Sequencing Method
- **C** Maxam-Gilbert Method
- **D** All of the above

1.1e The role of Urea in the sequencing gel is

- **A** to minimize DNA secondary structure which affects electrophoretic mobility
- **B** to decrease the viscosity of gel which affects electrophoretic mobility
- **C** To decrease the surface tension which affects electrophoretic mobility
- **D** All of the above

1.1f Which of the following is true for Sanger sequencing

- **A** ddNTPs are used, which lack the 3' hydroxyl group
- **B** DNA fragment to be sequenced are amplified by DNA polymerase and modified nucleotides are incorporated
- **C** single-stranded DNA molecules that differ in length by just a single nucleotide can be separated from one another by polyacrylamide gel electrophoresis
- **D** All the options are true

1.1g Pyrosequencing is an example of

- **A** Maxam-Gilbert Method
- **B** Chain Termination Method
- **C** Next-Generation Sequencing Method
- **D** Sanger Sequencing Method

1.1h nucleobase-specific partial chemical modification of DNA is used in:

- **A** Lambert Beer's Method

B NGS method
C Maxam Gilbert Method
D Pyrosequencing Method

1.1i Chain-terminating ddNTPs used in Sanger sequencing perform the chain termination by:

A 3'H prevents strand extension
B 5'P prevents strand extension
C 3'OH prevents strand extension
D All the options are correct

1.1j According to Clarke and carbon probability formula, if organism A has larger genome size than organism B (P=95% for both organisms); then minimum number of clones with 45 kb DNA insert size, to be screened will be:

A More for Organism A than B
B More for Organism B than A
C Equal for both the organisms A and B
D None of the options are correct

1.2 Answer the Following: (MCQ/Short Question/Fill in the Blanks) [05]

1.2a For 20 Kb insert, λ replacement vector can be used (T/F)

1.2b A vector digested with Sau3A enzyme can be ligated to an insert digested with XbaI, as the sticky end produced by both is same (T/F)

1.2c Full form of ddNTPs is_____

1.2d _____is a method to detect a polypeptide produced from a cloned gene

1.2e In nick translation, DNA to be processed is treated with _____to produce single-stranded nicks.

Q.2 Short Notes (Attempt any two) [06]

A DIG labelling
B Plaque hybridization
C Manual Sanger sequencing

Q.3 Explain in detail (Attempt any two) [14]

A What is high throughput sequencing? Explain two most widely used Next generation sequencing methods in detail
B Explain Maxam Gilbert Sequencing in detail?
C What do you understand by visual screening? Explain with three examples