(c) What are the advantages of cDNA library over a genomic DNA library?

3 + 6 + 3 = 12

7. (a) Describe different methods for size fractionation of gDNA.

(b) There is a protein G in a eukaryotic system, whose sequence of amino acid is known, describe the steps to clone the gene of protein G using a pUC18 vector.

6 + 6 = 12

Group – E

8. (a) Write names of three biopharmaceuticals. Describe the production of any one biopharmaceutical you mentioned through recombinant DNA technology with a diagram.

(b) Write names of three GM crop. Describe principle and steps for creation of golden rice producing transgenic paddy plant.

(c) (i) Define molecular biomarker with example? Write about the use of molecular biomarker in forensic science.

(ii) A researcher desires to clone a gene of a microorganism. Its genome size is 2.8×10^6 kb. The genomic library was created in vectors that were transformed into bacterial cells. If there is a 95% probability of the transformation, if he wants to screen 4.2×10^5 recombinant bacterial colonies to find this particular gene, then what will be average size of insert in this library?

(1 + 3) + (1 + 3) + (1 + 1 + 2) = 12

9. Write short notes about the following: **(3 × 4) = 12**

(i) Genetically engineered vaccine.
(ii) DNA based diagnosis of genetic diseases.
(iii) Human genome project.

B.TECH/BT/5TH SEM/BIOT 3103/2019

RECOMBINANT DNA TECHNOLOGY
(BIOT 3103)

Time Allotted : 3 hrs **Full Marks : 70**

Figures out of the right margin indicate full marks.

Candidates are required to answer Group A and any 5 (five) from Group B to E, taking at least one from each group.

Candidates are required to give answer in their own words as far as practicable.

Group – A
(Multiple Choice Type Questions)

1. Choose the correct alternative for the following: **10 × 1 = 10**

(i) Polygalactourinidase (PG) antisense RNA is used in reference to which phenomenon?
(a) Seed setting (b) Herbicide resistance
(c) Viral resistance (d) Fruit ripening.

(ii) Which of the following characteristic is undesirable for a vector?
(a) High copy number
(b) Ability to control their own replication
(c) Presence of several sites for a single restriction enzyme
(d) Small size.

(iii) The preferred enzyme used to remove 5' phosphate of DNA is
(a) BAP (b) CIAP
(c) SAP (d) Polynucleotide kinase.

(iv) Choose the correct sequence of events in a next generation sequencing technology-based whole genome sequencing project.
(a) DNA extraction, shearing, library preparation, sequencing, assembly, finishing, annotation, submission to Genbank.
(b) DNA extraction, library preparation, sequencing, assembly, finishing, annotation, submission to Genbank.
(c) DNA extraction, shearing, adapter ligation, library amplification, sequencing, assembly, finishing, annotation, submission to Genbank.
(d) DNA extraction, adapter ligation, library amplification, shearing, sequencing, assembly, finishing, annotation, submission to Genbank.

(v) Why are gene libraries constructed?
(a) To find new gene

(b) To sequence whole genome
(c) To create a "bank" of the genes in an organism
(d) All of the above.

(vi) A mixture contains three similarly sized peptides P, Q and R. The peptide P is positively charged, Q is weakly negative and R is strongly negative. If this mixture is passed through an ion-exchange chromatography column containing an anionic resin, their order of elution will be
(a) P, Q, R (b) R, Q, P
(c) Q, R, P (d) P, Q and R elute together.

(vii) Two plasmids are to be said compatible when
(a) they can co exist in the same bacterial cell
(b) carry the same antibiotic gene
(c) carry the same toxin gene
(d) all of the above.

(viii) A multimeric protein when run on an SD-PAGE, showed two bands at 20 KDa and 40 KDa. However, when the protein was run on a native PAGE, it showed a single band at 120 KDa. The native form of the protein would be
(a) homotrimer (b) heterotetramer
(c) heterodimer (d) heterotrimer.

(ix) Pure plasmid DNA was isolated from a bacterium. Restriction enzyme digestion of this plasmid with either *Bam* HI or *Eco* RI resulted in two DNA fragments. A double digestion of the same plasmid with both these enzymes resulted in three DNA fragments. From this we can conclude that the isolated plasmid DNA is
(a) Double stranded and linear (b) Double stranded and circular
(c) Single stranded and linear (d) Single stranded and circular.

(x) A contig is a
(a) map of genetic markers that separated by less than 1cm
(b) map showing the order of cloned bits of DNA
(c) unique DNA sequences that serve as molecular markers
(d) set of two or more partially overlapping cloned DNA fragments.

Group – B

2. (a) How pBR322 was designed?

(b) What is replica plating method?

(c) Discuss the selection of recombinants by this method with a labelled diagram.

3 + 3 + 6 = 12

3. (a) What do you understand by the terms isoschizomers, neoschizomers, isocaudomers? Give examples of each highlighting the advantages of each in gene cloning.

(b) Suppose we have isolated the gene of interest by cleavage with Bam HI, and we have cleaved the vector with Bgl II, can we ligate the two without the linkers or adaptors? If yes, can the hybrid site (Bam HI/ Bgl II) be recleaved with any of the enzymes? Explain.

(c) What are linkers and adapters? Mention their different applications.

5 + 3 + 4 = 12

Group – C

4. (a) Why we prefer nonradioactive probe instead of radioactive probe? Explain with diagram that how you will identify a DNA from a mixture of DNA using DIG labelled ss-DNA probe using a chemiluminescent substrate.

(b) Write names of three techniques for identification of DNA, which are based on principle of fluorescence. Describe any one of that techniques which is *in vivo*.

(c) Three restriction endonucleases (RE-A, RE-B and RE-C) are used to cut a piece of circular DNA, singly and in pair wise combination. From the agarose gel electrophoresis result, sizes of fragments (in kb) are listed in order of size. Determine the correct order of restriction sites, and draw the final restriction map of the DNA, with the intervals between sites labelled. A) 17; B) 17; C) 10, 7; A x B) 12, 5; A x C) 10, 6, 1; B x C) 7, 6, 4.

(d) Write reaction with enzyme and condition for the following conversion:
(i) Blunt end ds DNA to staggered end ds DNA
(ii) From a ssRNA to ssDNA.

(1 + 3) + 3 + 3 + 2 = 12

5. (a) Explain the principle of Automated PCR cycle sequencing of DNA.

(b) What is ELISA? Write names of different types of ELISA. Explain principle of detection of antigen by any one type of ELISA.

(c) Explain the principle of purification of protein with His-tag from *E.coli* extract.

3 + (1 + 1 + 3) + 4 = 12

Group – D

6. (a) What is cDNA library?

(b) Describe preparation of cDNA library by schematic diagram.