KATHMANDU UNIVERSITY
End Semester Examination
July/August, 2024

Marks Scored:

Level : B.Tech.
Year : III

Course : BIOT 306
Semester : II

Exam Roll No. :

Time: 30 mins.

F. M. : 20

Registration No.:

Date : 30 JUL 2024

SECTION "A"

[20 Q. × 0.75 = 15 marks]

Choose and mark [X] in the most appropriate option from each set of choices

1. "Naked" DNA is.
   [ ] free of cells
   [ ] free of nucleic acids
   [ ] free of proteins
   [ ] contains just the sugar-phosphate backbone

2. _____ consists of recombinant cells containing different fragments of a foreign genome.
   [ ] Homologous recombinants
   [ ] Genomic libraries
   [ ] DNA probes
   [ ] Knockout organisms

3. Gene targeting is done on a(n)
   [ ] sperm cell
   [ ] early embryonic cell
   [ ] egg cell
   [ ] fertilized ovum

4. If you are using a vector with lacZ as a selectable marker, which of the following bacterial strains would you want to use as host cells?
   [ ] Lac O^- [ ] Lac I^- [ ] Lac Z^- [ ] Lac Z^+

5. What key feature of *Taq* polymerase allows PCR to be conveniently performed?
   [ ] *Taq* polymerase does not require a primer
   [ ] *Taq* polymerase does not require a template
   [ ] *Taq* polymerase is not damaged by heating
   [ ] *Taq* polymerase can work at very low temperature

6. The exonuclease enzyme that removes nucleotides from both the strands of a DNA.
   [ ] Exonuclease III [ ] S1 [ ] Bal31 [ ] DNaseI

7. The strain of *E. coli* used in single-strain in-vitro packaging system.
   [ ] BHB2688 [ ] BHB2690 [ ] SMR09 [ ] SMR10

8. Location of transcription start point can be done by
   [ ] Northern hybridization
   [ ] S1 nuclease treatment
   [ ] Primer extension
   [ ] Electron microscopy

9. The vector used for the construction of antisense polygalacturonase gene is
   [ ] pBIN16 [ ] pBIN17 [ ] pBIN18 [ ] pBIN19

10. In insertional inactivation of SUP4 the recombinants are colored.
[ ] White [ ] Red [ ] Yellow [ ] Blue

11. Although the Ti plasmid has revolutionized plant genetic engineering, one limitation of its use is that it
[ ] Cannot infect broadleaf plants
[ ] Cannot be used on fruit-bearing plants
[ ] Cannot transmit prokaryotic genes
[ ] Does not infect cereal plants such as corn and rice

12. For estimating the cell number of a bacterial cell optical density reading is taken at
[ ] 260nm [ ] 280nm [ ] 600nm [ ] 660nm

13. Cultured animal cells are transformed, especially if the DNA is precipitated onto the cell surface with
[ ] Sodium chloride [ ] Calcium phosphate
[ ] Calcium chloride [ ] Potassium phosphate

14. Both the restriction enzymes produce GATC sticky ends.
[ ] BamHI and BglII [ ] PvuI and PvuII
[ ] HindIII and HinfI [ ] AluI and TaqI

15. The most common type of STR found in human genome is
[ ] TA [ ] TC [ ] CA [ ] GA

16. The glucoamylase gene is induced by
[ ] Galactose [ ] Starch [ ] Cellulose [ ] Methanol

17. Protein factor VIII is used in the treatment of
[ ] Diabetes [ ] Haemophilia [ ] Anaemia [ ] Emphysema

18. GLYAT detoxifies glyphosate by attaching _____ group to the herbicide molecule.
[ ] Methyl [ ] Acetyl [ ] Amine [ ] Keto

19. The first breakthrough in mapping the human breast cancer susceptibility gene BRCA1 occurred in 1990 as a result of _____ analyses.
[ ] PCR [ ] RFLP [ ] STR [ ] RAPD

20. The number of amino acids present in B chain of insulin molecule.
[ ] 19 [ ] 22 [ ] 27 [ ] 30

SECTION "B"
[10 Q. × 0.5 = 5 marks]

Fill in the blanks

21. Conjugal transfer of plasmids is promoted by _____ plasmids.

22. _____ is the enzyme involved in the digestion of polymeric compounds of the cell wall of *E.coli.*

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23. _____ is the example of non-ionic detergent.

24. In order to use LEU2 gene as a selectable marker the host must be _____ mutant.

25. The targeted gene for gene subtraction projects which leads to reduction of starch content in vegetables is _____.

26. _____ is the viral vector which is used in insects for the production of recombinant protein.

27. Sex identification by PCR of part of the amelogenin gene produces_____ band in males.

28. In a technique called virus-induced gene silencing the viral _____ is degraded.

29. If the protein does not take up its correctly folded tertiary structure, then usually it is insoluble and forms _____within the bacterium.

30. For the production of recombinant protein in milk the gene is attached to promoter of _____ gene.

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SECTION "C"
[3 Q. × 7 = 21 marks]

Attempt *ANY THREE* questions.

1. What is DNA sequencing? Describe the reversible terminator sequencing method of NGS sequencing in brief. [2+5]

2. What is the importance of restriction mapping in genetic engineering? A linear λ DNA molecule when restricted with EcoR1 produces two fragments of size 22.0 and 26.5. The same DNA when restricted with another BamH1 also produces two fragments of size 13.0 and 35.5. Double digestion however yields fragments of size 9.0, 13.0 and 26.5. Construct restriction map of the DNA using the information provided showing restriction sites of respective enzymes. [2+5]

3. Write down the basic principle of a PCR reaction. How real time quantitative PCR is different from conventional PCR? Elaborate. [2+5]

4. Describe the structure of 2μm plasmid. What strategies are used for cloning genes in yeast artificial chromosome? [2+5]

SECTION "D"
[34 marks]

5. Write short notes on (***ANY FOUR***): [4 Q. × 3.5 = 14]
   a. Problems with production of recombinant proteins in *E. coli*.
   b. Terminator technology.
   c. *In vitro* mutagenesis techniques.
   d. Ion-exchange chromatography for the purification of DNA.
   e. Lac selection.

6. Write down **TWO** differences between (***ANY FOUR***): [4 Q. × 2 = 8]
   a. Hybrid-release translation (HRT) and hybrid-arrest translation (HART).
   b. Conjugation and Compatibility.
   c. Cosmid and Phagemid.
   d. Linkers and Adaptors.
   e. P- element and SV40 virus.

7. Give reasons **WHY/ HOW**? (***ANY FOUR***): [4 Q. × 3 =12]
   a. A lower than optimal PCR annealing temperature leads to non-specific product synthesis.
   b. Cloning **δ**-endotoxin genes in chloroplasts is preferred.
   c. Production of recombinant somatotrophin in *E. coli* is done.
   d. pUC8 vectors are considered more efficient than pBR322 vectors.
   e. DNA molecule is first covered with protein molecule before observing under electron microscope.