

18. i) How does quadrupole analyze the mass of a peptide in a mass spectrometry? 4
- ii) What kind of ionization would you choose for volatile and non-volatile substances? 2
- iii) What is the basic principles of separation of proteins in 2D-GEL electrophoresis? Describe how would you perform isoelectric focusing gel electrophoresis in order to separate proteins? 1+3=4

**M.Sc. (BIOTECHNOLOGY) PART - II EXAMINATION, 2019**

**MOLECULAR GENETICS, GENOMICS AND PROTEOMICS**

**MSBT 2/2**

Time : Four hours

Full Marks : 100

**PART - I**

**GROUP - A**

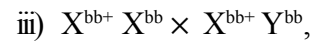
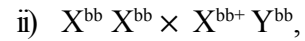
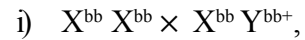
Answer question no. *1* and *any two* from the rest 10×3

1. a) How the term “incomplete-dominance” differs from “co-dominance” at the molecular level? Explain with an example. 2
- b) Briefly explain what is a ‘Molecular Marker’? How the molecular marker can be used to make genetic map? 2
- c) How the disease ‘phenylketonuria’ (PKU) is caused? What kind of lesson we learnt from the distribution of mutant alleles on the gene of interest causing this disease. 2
- d) “Despite having a similar composition of the sex chromosomes in the males and females of both *Drosophila* and human, the mechanism of sex determination in these two organism is entirely different”. Justify this statement. 2
- e) What is ‘lod’ score? Explain how this parameter can be used to assess the linkage in human pedigrees? 2

[ Turn over

[ 2 ]

2. a) In *Drosophila*, the gene for bobbed bristle (recessive allele  $bb$ , bobbed bristles ; wild type alleles  $bb^+$  normal bristles) is located on the X chromosome and on a homologous segment on the Y chromosome. Give the genotypes and phenotypes of the offspring from the following crosses :



b) Explain what do you mean by “complementation group”? In an attempt to identify the genes involved in leucine biosynthesis in *Neurospora* you have initially mutagenized a wild type strain by methyl methane sulphonate. Initially you have obtained 745 strains who failed to grow on minimal media lacking leucine, but all of them have grown fine on rich YPD medium. Outline an approach how you would ascertain if all 745 leucine-auxotrophic mutant strains belong to one class. If not, how you would classify them experimentally using systematic genetic analysis. 4

c) What do you mean by “penetrance” and “suppression”. Provide examples. 2

[ 13 ]

V	99·1326	M	131·1986
T	101·1051	H	137·1412
C	103·1448	F	147·1766
I	113·1595	R	156·1876
L	113·1595	Y	163·1760
N	114·1039	W	186·2133

16. i) What are the basic components required for a mass spectrometry? 2

ii) What does MALDI-TOF stand for? Explain briefly the basis of this technique for the analysis of proteins? 1+5=6

iii) What is the function of electron multiplier in mass spectrometry? 2

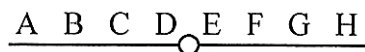
17. i) Explain briefly the method of cell labelling by SILAC for quantitative proteomic analysis. 2

ii) What are the advantages of using SILAC over stable isotope based labelling for quantitative proteomic analysis? 2

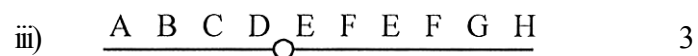
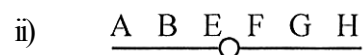
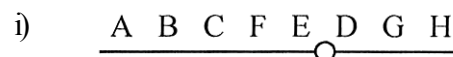
iii) What are molecular ions, fragment ions and base peak in mass spectrometric analysis? 2×3=6

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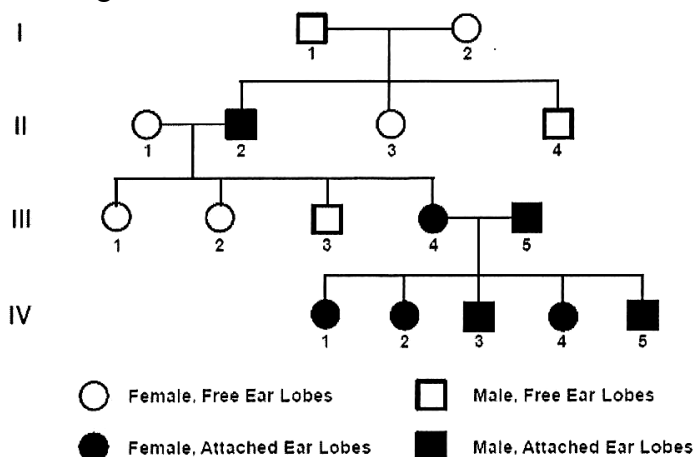
- b) Normal Chromosome of a certain organism has the following configuration of genes :



One member of this organism was irradiated with UV-radiation and crossed with the opposite mating type having recessive alleles for all these genes. In the progeny of the cross organism with following types of genotypic configuration were obtained in addition to individuals with normal configuration. Account for the observed results :



4. a) The following is a pedigree of ear lobe shape in human beings :



- c) You have identified a new protein in *Saccharomyces cerevisiae*. A preliminary bioinformatic analysis is indicating that this protein could be transcription factor and posses a putative leucin zipper domain and a stretch of poly-glutamine towards the c-terminal end of the primary sequence. Suggest an experiment to identify the putative target genes of this newly identified transcription factor in genome wide scale. Briefly explain the rationale of such procedure. 5

14. a) What do mean by “Genome Sequencing by ordered clone approach”? 2

- b) You would like to analyze and compare the transcriptome of primary mammary epithelial cell line and a cell line, Hs-274.T that is derived from breast a cancer patient. After comparing them you have found almost 1570 genes which were upregulated in the tumour line and 377 genes which were down-regulated. Suggest a control experiment, which would justify the validity of your finding with appropriate reasoning. 3

- c) Briefly explain in principle of ‘Bridge Amplification’ reaction with necessary diagram. 3

- d) Explain the rationale of the ‘sequencing by ligation’ procedure. 2

[ 6 ]

Explain the MspII digestion data and provide the genotype (with respect to Sickle cell anemia) of each of the family member. 6

- b) Provide an evidence that crossing over involves breakage and rejoining of strands of two non-sister homologous chromosome. 4

### GROUP - B

Answer *any three* questions 3×10

6. a) In an experiment in conjugation, the donor strain is streptomycin sensitive, so that it can be eliminated by growing the exconjugants in medium containing streptomycin. At which end of the chromosome would you want the streptomycin sensitive gene to be located - at the origin of transfer or at the distal end? 5
- b) Why P1 phage can not, but phage  $\lambda$  can be recovered from the recipient cell upon transduction of the genes from the donor to the recipient? 5
7. a) In an interrupted mating experiment, five *Hfr* strains were tested for the sequence in which they transmitted ten gene markers F, G, O, P, Q, R, S, W, X, Y to an  $F^-$  cell. The order of transmission were :
- i) Q S R P O F
- ii) Y G F O P R

[ 9 ]

- b) Design an experiment to determine whether a particular drug really causes mutations or merely identifies mutations which are already present in the organism under investigation. 5

### GROUP - C

Answer *any two* questions

11. a) You have the following sequence reads from a genomic clone of the *Homo sapiens* genome :

Read 1 : ATGCGATCTGTGAGCCGAGTCTTTA

Read 2 : AACAAAAATGTTGTTATTTTTATTTCAGATG

Read 3 : TTCAGATGCGATCTGTGAGCCGAG

Read 4 : TGTCTGCCATTCTTAAAAACAAAAATGT

Read 5 : TGTTATTTTTATTTCAGATGCGA

Read 6 : AACAAAAATGTTGTTATT

Use these six sequence reads to create a sequence contig of this part of the *H. sapiens* genome. 4

- b) You have sequenced the genome of the bacterium *Salmonella typhimurium*, and you are using BLAST analysis to identify similarities within the *S. typhimurium* genome to known proteins. You find a protein that is 100 percent identical in the bacterium *Escherichia coli*. When

[ Turn over

experimental precaution following the UV irradiation is normally taken to maximize the effect of UV irradiation ?

3

- c) Two different cells of yeast *S. cerevisiae* have undergone two different sets of mutations in the same gene encoding an essential growth function following mutagenesis event. One cell contains a base substitution mutation while the other one has a single nucleotide deletion. Explain with reasoning which cell is expected to suffer more lethal effect. 2
- d) What is main source of spontaneous mutations in a cell ? Are mutation and polymorphism synonymous ? Explain with reasoning. 2
9. a) How the lysogeny is established in Phage  $\lambda$ . Describe the series of events with the necessary diagrams. 6
- b) Are the linkage maps of the mature phage chromosome (vegetative map) and that of the  $\lambda$  prophage identical ? If not, how do they differ ? Explain which significant conclusion about the state of phage DNA during lysogenization was deduced from this observation. 4
10. a) Explain with necessary diagram how Benzer deduced that rII locus of phage T4 is composed of two cistrons. 5

iii) R S Q W X Y

iv) O P R S Q W

v) Q W X Y G F

What is the gene sequence in the original strain from which the *Hfr* strain were derived ?

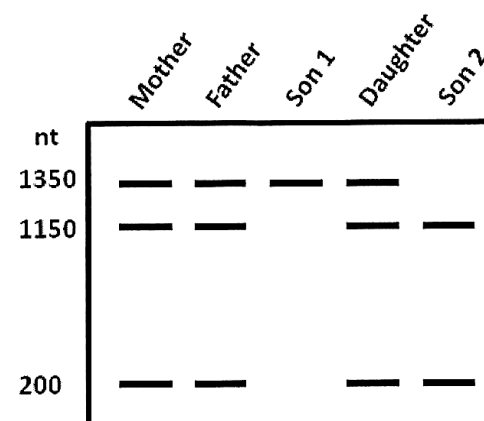
For each *Hfr* strain, state which donor gene marker should be selected in the recipients after conjugation to obtain the highest proportion of recombinants that will be *Hfr*? 3+2

- b) What are the features of lambda and P1 phage that make one a restricted and the other an unrestricted transducing phage ? 3
- c) If you use phage  $\lambda$  in Luria-Delbruck fluctuation Experiment, what would have been the consequences ? 2
8. a) If you want to isolate a replication dependent mutant strain of *E. coli*, how would you isolate such a mutant strain ? (Recall that a strain defective in replication may not be viable). 3
- b) Typically what kind of mutation is induced by treatment of the cell by high dose of UV radiation ? What kind of

you compare nucleotide sequences of the *S. typhimurium* and *E. coli* genes, you find that their nucleotide sequences are only 87 percent identical.

- i) Explain this observation.
  - ii) What do these observations tell you about the merits of nucleotide versus protein similarity searches in identifying related genes? 6
12. a) What is the meaning of “base-calling” in next-generation sequencing platforms? Explain the principle of reversible terminator technology of DNA sequencing used by Illumina Genome Analyzer. 2+4
- b) Why is it necessary to validate the expression dataset obtained from microarray analysis performed at the “transcriptomic” scale. Mention two methods which are routinely used to validate the expression datasets. 2+2
13. a) A global analysis of gene expression can be accomplished by using a DNA microarray. What is a DNA microarray? How DNA microarrays are used for studying gene expression? 3
- b) What do you mean by “probe set” in a microarray experiment. Briefly explain why a combination of perfectly matched and a mismatched oligonucleotide is used to design such a probe set. 2

- i) State which pattern of inheritance for this trait is consistent with this pedigree. Give reasons.
  - ii) Using the first letter of the alphabet, give the genotype of each of the following persons :  
I-1, II-1 and III-4
  - iii) No ancestry information is available for II-1. How do you justify the assignment of her genotype.  $3 \times 2 = 6$
- b) Briefly describe the experimental outline of Beedle and Tatum’s experiment with *Neurospora crasa*. What fundamental principle of genetics they have coined from this experiment? What is the implication of this principle in modern biology? 2+1+1
5. a) Sickle cell anemia is a recessive genetic disease caused due to a point mutation in the 6th codon abolishing one of the MspII endonuclease digestion site present in  $\beta$ -globin gene. MspII digested DNA from a normal person gives two bands, 1150 bp and 200 in  $\beta$ -globin gene. The MspII digestion pattern of  $\beta$ -globin gene from the blood samples from the members of a family is given below :



**PART - II**

Proteomics (Answer **any two** each containing 10 marks, Total 20)  
10×2

15. i) What is the purpose of digesting the protein with trypsin for mass spectrometric analysis? 2
- ii) Why trypsin is mostly used over pepsin to digest protein for mass spectrometric analysis? 2
- iii) Can the following two peptides with the same amino acid composition be distinguished by MALDI-TOF mass spectrometry? Explain. Can these peptides be distinguished after digestion with trypsin? 1+1=2

**Peptide A : GASPVRTCILKMHFY**

**Peptide B : MGHFRAITKYPVCSL**

- iv) Calculate the expected (monoisotopic) masses if the enzyme trypsin was used to digest peptides A and B. [A table of amino acid masses is shown below]. 4

Amino acid	Mass(avg)	Amino acid	Mass(avg)
G	57.0520	D	115.0886
A	71.0788	Q	128.1308
S	87.0782	K	128.1742
P	97.1167	E	129.1155

3. a) Following three recessive genes are found in corn : bt, brittle endosperm ;  $g^{117}$ , glossy leaf ; rg, ragged seedling. A trihybrid of unknown origin is test crossed, producing the following offspring :

<u>Phenotypes</u>	<u>Number</u>
Brittle, glossy, ragged	236
Brittle, glossy	241
Ragged	219
Glossy	23
Wild type	224
Brittle ragged	17
Glossy ragged	21
Brittle	19
Total	1000

- i) If the genes are linked, determine the relative order and map distance.
- ii) Reconstruct chromosome of trihybrid.
- iii) Is there any crossover interference? If yes, how much. 4+1+2=7