Name : .....

Roll No. : .....

Invigilator's Signature : .....

CS/M.TECH(BT)/SEM-2/MBT-215D/2012

2012
PROTEOMICS AND GENOMICS

Time Allotted : 3 Hours *Full Marks* : 70

The figures in the margin indicate full marks.
Candidates are required to give their answers in their own words as far as practicable.

GROUP – A

(Multiple Choice Type Questions)

1. Choose the correct alternatives for any *ten* of the following : $10 \times 1 = 10$

i) For a coenzyme binding with a ligand like NAD, K_d, the dissociation constant varies between

a) $10^{-4} - 10^{-2}$ b) 10^{-7} to 10^{-4}

c) 10^{-4} to 10^{-11} d) 10^{-15}.

ii) The 2D-Gel Electrophoresis provide information's about the proteins are

a) MW, pI and quantity b) MW and pI

c) pI and quantity d) none of these.

iii) Protein visualization sensitivity by Coomassie Brilliant Blue R-250 Staining per spot is

a) 1 ng to 1 μg b) 100 ng to 10 μg,

c) 1 ng and up d) 1 mg.

iv) Which of the following techniques DO NOT measure protein-protein and protein-nucleic acid interactions directly ?

a) X-ray and NMR structure determination

b) Chemical cross-linking

c) Phage Display

d) UV-spectrophotometry.

v) Trypsin cleave the peptide bond containing

a) Arg or Lys

b) Glu or Asp

c) Met, Trp.

vi) The whole genome shotgun sequencing was first commercially used by

a) Craig Venter b) Francis Collins

c) James Watson d) Eugene Myers.

vii) GFP tagging & microscopy is used in proteomics to

a) investigate protein-protein interaction,

b) identify the protein

c) find out the localization of the protein

d) study the structure of the protein.

viii) The lack of evolutionary divergence in ubiquitin from different organisms suggests

a) most residues of ubiquitin have essential role

b) functional importance of ubiquitin

c) has a key role in the pathway of degradation

d) all of these.

ix) To study peptides, proteins and DNA up to 500 kD by mass spectrometry which ionization method will be the best ?

a) Electron impact ionization

b) ESI

c) MALDI

d) FAB.

x) The amino acid composition of protein-protein interfaces are maximally composed of which of the following residue sets ?

a) His-Phe-Trp-Tyr b) Lys-Arg

c) Leu-Ile d) Val-Met.

xi) An overlapping series of clones or sequence reads that corresponds to a contiguous segment of source genome is called a

a) clone library b) contig

c) snip-SNP d) STS.

xii) In human mitochondrial DNA, heavy strand is rich in

a) guanine b) cytosine

c) adenine d) uracil.

GROUP – B

(Short Answer Type Questions)

Answer any *three* of the following. $3 \times 5 = 15$

2. Write the principle, application and describe with diagram about any one of the following tools :

   i) Affinity pull down assay

   ii) phage display

   iii) Yeast two hybrid

   iv) Protein microarray.

3. List three important parameters each in NMR spectroscopy and X-ray crystallography that are of importance for biological macromolecule structure determination. Briefly cite the reasons why they are important.

4. a) How can FRET be used for measuring protein-protein interactions directly ?

   b) For avidin-biotin, $K_D = 10^{-15}$. If k_{on} were 10^{-7} $M^{-1}s^{-1}$ what would be the half life of the complex ? (assume diffusion limit to be 10^{-9} $M^{-1}s^{-1}$) 2 + 3

5. Mention the salient features of the human mitochondrial genome with a diagram.

6. Briefly describe the principle of Pyrosequencing with a flow diagram.

7. Mention the different methods of hybridization and role of temperature and Na^+ in hybridization procedure in microarray. 3 + 2

8. In Eukaryotic system of gene prediction programs *ab-initio* based programmes rely on the several features. Describe.

GROUP – C

(Long Answer Type Questions)

Answer any *three* of the following. $3 \times 15 = 45$

9. Elucidate the idea of Hydrogen hypothesis with respect to genome evolution. How did exon duplication and exon shuffling contribute to formation of new genes ? Give a comparative account of functionally similar genes, functionally identical genes and functionally related genes.

5 + 5 + 5

10. a) What is proteome ?

    b) What is protein ? What is the life cycle of protein ? (Write with a diagram)

    c) What are the basic differences between proteomics and protein chemistry ?

    d) What are the different types of proteomics and the tools and techniques use to study these ?

    e) What are the needs of expressional proteomics ?

1 + 4 + 3 + 4 + 3

11. a) Use 2 examples from the protein world to highlight the importance measuring lifetimes of protein complexes.

    b) How is complementarity of interfaces important for specificity of a protein-protein interaction ? Use an example to point out this specificity.

    c) For a protein-protein interface of 1700 A^2, what should be the minimum surface area ? If the entire area were hydrophobic, what contribution to the free energy of stabilization would be made ? Show calculation.

    d) Briefly use the concept of 'surface footprinting' to explain the organization of protein complexes in 3 dimensions. 3 + 4 + 4 + 4

12. Discuss the basic principle behind the AFLP technique. "A combination of shotgun sequencing and physical mapping now is the favored method for sequencing large genomes". Justify the statement with respect to human genome sequencing. What do you mean by genes within genes ? What is a gene superfamily ? 4 + 5 + 3 + 3

13. a) What are the disadvantages of 2-D PAGE ?

    b) Describe the basic principles of ESI-MS.

    c) Write the advantages of MALDI-M over ESI-MS.

    d) A protein from human tissue Mol. WT. determined ~ 12,000 by SEC and 13, 000 by SDS-PAGE. Pure protein subjected to ESI-MS and following data obtained.

m/z	773.3	825.5	884.3	952.3	1031.3
Abundance (%)	59	88	100	66	37

    Given that $n_2 = (m_1 - 1)/(m_2 - m_1)$ and $M = n_2 (m_2 - 1)$ and assuming that the only ions in the mixture arise by protonation, deduce ave. mol. Mass for protein by this method. 5 + 3 + 4 + 3

14. a) Draw a schematic for the ubiquitin-mediated degradation of cytosolic proteins.

    b) Briefly outline the PEST mechanism of protein turnover highlighting what is still unknown about the structural basis for this increased turnover rate.

    c) Use a representative data set from 4 organisms (e.g. H phlori, C.elegans, D.melanogaster and S.cerevisiae) to highlight how a binary protein interaction network can be detected by a 'High throughput' – two hybrid screening systems. 4 + 5 + 6

15. a) What is microarray ?

    b) Mention the steps of a typical DNA microarray experiment.

    c) Mention the steps of feature extraction.

    d) Mention names of any two softwares which are used for image processing.

    e) Describe the different methods of within array normalization with special reference on two-color microarray.

    f) Mention the applications of microarray 2 + 3 + 2 + 5 + 3

16. Write short notes on any *three* of the following. 3 × 5

    a) BLAST steps

    b) Extrinsic method of gene prediction

    c) Prokaryotic gene prediction features

    d) Fitch-Margoliash method.