(b) If two analytes have different molecular masses and Kd values, what is the difference in their elution volumes?

(c) The M_r (relative molecular mass) of a *unknown* protein was being investigated by SE chromatography with Sephacryl S300 column using the enzymes aldolase, catalase, ferritin, thyroglobulin and Blue Dextran as standards. The M_rs and the retention volumes (V_rs) of the standards are known as is the retention volume of the unknown protein. *How would you deduce the relative molecular mass of the unknown protein*?

(d) Name six important parameters for the development of a successful immunoassay.

(e) What makes fluorescence a good detection technique for immunoassay development? Draw a flowchart to represent the development, optimization and validation of a immunoassay.

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

Group - E

8. (a) Explain the characteristics of Non-invasive biosensors.

(b) How can you detect DNA with the help of optical biosensor ?

6 + 6 = 12

9. (a) Name the three enzymes whose levels in blood plasma are typically affected following a myocardial infarction. Draw a plot to explain the differences in the time courses of release/disappearance of these enzymes into the plasma. Itemize the factors that make these time courses different and explain how they have been exploited for clinical diagnostics purposes.

(b) Draw a labeled diagram (with biochemical reaction) for an amperometric glucose biosensor that is based on enzymatic conversion of glucose to hydrogen peroxide. How has *carbon nanotube technology* been utilized to make a glucose micro-biosensor? Itemize its operating principles

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

B.TECH/BT/8TH SEM/BIOT 4246/2018

MEDICAL & PHARMACEUTICAL BIOTECHNOLOGY
(BIOT 4246)

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) The alpha-receptor polypeptide constituent of the high affinity human IL-2 receptor has a molecular mass of
(a) 55 kDa (b) 75kDa (c) 64kDa (d) 82 kDa.

(ii) First liver enzyme to rise in the blood when the bile duct gets obstructed is
(a) Alkaline phosphatase (b) Lactate dehydrogenase
(c) Gamma-glutamyltransferase (d) Alanine transaminase

(iii) Diagnostic fluids recognized by non–invasive biosensor is
(a) sweat (b) saliva
(c) blood (d) both (a) & (b).

(iv) A carrier for a peptide vaccine maybe one that is used for the vaccination itself. An example of such a carrier is
(a) influenza hemaglutinin (b) tetanus toxoid
(c) plasmodium falciparum (d) tobacco mosaic virus.

(v) For human insulin prb, the final purification step involves
(a) cell recovery and homogenization
(b) RP-HPLC
(c) ion exchange chromatography
(d) gel filtration chromatography.

(vi) Warfarin, a coumarin derivative, is therapeutically used as an
(a) antipyretic (b) analgesic
(c) anti-coagulant (d) anti-hypertensive.

(vii) "Single peak insulin" is
(a) a recombinant human insulin that has a long lasting response
(b) bovine or porcine insulin that has undergone an additional gel filtration purification step
(c) recombinant human insulin produced in yeast
(d) insulin purified by gel filtration and ion exchange chromatography.

(viii) Addition of β-mercapto ethanol in SDS-PAGE causes
(a) disruption of inter-chain disulfide linkages
(b) disruption of intrachain disulfide linkages
(c) same effect as dithiothreitol
(d) all of the above.

(ix) Reverse phase HPLC separates proteins on the basis of differences in
(a) molecular mass (b) fractionation speed
(c) surface hydrophobicity (d) charge.

(x) For combination immunotherapy, which of the following agents can be combined with Interleukin-15 administration?
(a) A vaccine (b) A checkpoint inhibitor
(c) An *adoptively* transferred T-cell (d) None of the above.

Group – B

2. (a) Itemize the major tests that are undertaken on a potential new drug during the preclinical trial phase of drug development. What factors are emphasized upon in the preclinical phase? Choose *any* TWO characteristic tests done during this phase of drug development and explain *the difference in these two characteristics* between a small molecule drug and a biopharmaceutical.

(b) If one measures the pharmacokinetic profile of a drug, what are the relevant parameters? Explain the parameters with relevant mathematical expressions*, where applicable*.

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

3. (a) Draw a flow chart showing the steps for a likely *high fidelity* purification of recombinant therapeutic insulin. What is the necessity of a possible RP-HPLC based last step? Could you suggest a different chromatographic route for this step? *Explain your answer*.

(b) A drug like the antibiotic vancomycin can exhibit a slow equilibration time with its redistribution in peripheral tissues. *Draw a two-compartment model to represent this pharmacokinetic*

behavior. What would a plot of log C_p vs time for such a 2 compartment model look like?

(c) In analyzing a flowchart showing the preparative steps of a rh-EPO procedure, there are 4 chromatographic steps. What chromatographic techniques are utilized? Cite the reasons for their usage. Citing examples, explain the reasons for shift in therapeutic usage of rh-EPO to non-renal indications.

(d) Using a labeled diagram, explain how the principles of *rational* drug design have been utilized in the production of synthetic betamethasone.

3 + 2 + (2 × 2) + 3 = 12

Group – C

4. (a) Give a concise definition of pharmacogenomics. In simple terms, explain how has this sub-discipline influenced medical treatment? How does the polymorphism of CYP genes affect drug metabolism?

(b) Explain the physiological mechanism behind the action of a DNA vaccine.

(1 + 2 + 4) + 5 = 12

5. (a) Discuss the applications of Monoclonal antibodies.

(b) How can a specific gene expression be inhibited with the help of Peptide Nucleic Acid (PNA)?

6 + 6 = 12

Group – D

6. (a) How can proteomics be utilized to assess drug toxicity during clinical development? Define a toxicity biomarker. What techniques have been utilized to discover toxicity biomarkers? Using one example highlight the working principle of a toxicity biomarker.

(b) How does capillary electrophoresis (CE) play an important role in the quality control phase of a biopharmaceutical's production ? What are its key benefits in so far as separation technology principles are concerned?

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

7. (a) Schematically represent how proteins of different sizes are separated by size exclusion chromatography.