QUESTION BANK
Academic Year 2019-2020

Class	: M. Sc. I (Semester- I)
Paper Code	: **MICRO4202**
Paper	: II
Title of Paper	: **Instrumentation**
Credit	: 4
No. of lectures	: 60

Questions for 2marks

1. Define the following.
   a. Partition coefficient
   b. Resolution (in context of chromatography)
   c. Electro-magnetic radiation
   d. Atomic orbitals
2. What is Beer's law?
3. What is Lambert's law?
4. In which type of chromatography, the stationary phase held in a narrow tube and the mobile phase is forced through it under pressure?
   a. Column chromatography
   b. Planar chromatography
   c. Liquid chromatography
   d. Gas chromatography

5. In chromatography, the stationary phase can be _____ supported on a solid.
   a) Solid or liquid
   b) Liquid or gas
   c) Solid only
   d) Liquid only

6. In chromatography, which of the following can the mobile phase be made of?
   a) Solid or liquid
   b) Liquid or gas
   c) Gas only
   d) Liquid only

7. Which of the following types of chromatography involves the separation of substances in a mixture over a 0.2mm thick layer of an adsorbent?
   a) Gas liquid
   b) Column
   c) Thin layer
   d) Paper

8. Chromatography cannot be used to purify volatile substances.
   a) True
   b) False

9. Gas chromatography can be performed in which of the following ways?
   a) Only in columns
   b) Only on plane surfaces
   c) Either in columns or on plane surfaces
   d) Neither in columns nor on plane surfaces

10. In Gas-liquid phase chromatography, the stationary phase is composed of _____ and the mobile phase is made of _____
    a) Solid, liquid
    b) Liquid, liquid
    c) Liquid, gas
    d) Solid, gas

11. Which of the following types of chromatography involves the process, where the mobile phase moves through the stationary phase by the influence of gravity or capillary action?
    a) Column Chromatography
    b) High Pressure Liquid Chromatography
    c) Gas Chromatography
    d) Planar Chromatography

12. Pattern on paper in chromatography is called
    A. chroming
    B. chroma
    C. chromatograph
    D. chromatogram

13. Ion exchange chromatography is based on the
    A. electrostatic attraction
    B. electrical mobility of ionic species
    C. adsorption chromatography
    D. partition chromatography

14. Explain the principle of chromatography.
15. Enlist the detectors used in HPLC.
16. Enlist the detectors used in GC.
17. Explain the difference between planer and column chromatography.
18. Give the principle of GC.
19. Explain partition coefficient.
20. Give the van Deemter equation.
21. Give principle of Gel filtration chromatography.
22. Give principle of Ion exchange chromatography.
23. Give principle of Affinity chromatography.

Questions for 4 marks

1. Describe the working of SDS-PAGE.
2. Enlist the different methods for purifying proteins and explain anyone with a suitable

example.

3. Describe ultracentrifugation with a suitable diagram.
4. Differentiate between TEM and SEM.
5. Explain the principle of Gel filtration chromatography.
6. Explain in brief isoelectric focusing.
7. Explain the instrumentation of HPLC with a suitable diagram.
8. Differentiate between Isopycnic and Rate zonal centrifugation.
9. Describe the principle of isoelectric focusing.
10. Differentiate between bathochromic and hypsochromic shift.
11. Write a short note on the exchangers used in ion exchange chromatography.
12. Describe the components of fluorescence spectrophotometer.
13. Explain the principle of affinity chromatography.
14. Write a short note on Gas chromatography.
15. Write a short note onuv/visible spectroscopy.
16. Write a short note on IR spectroscopy.
17. Write a short note on circular dichroism.
18. Write a short note onagarose gel electrophoresis.
19. Write a short note on native PAGE.
20. Write a short note on confocal microscopy.
21. Write a short note on phase contrast microscopy.
22. Write a short note on Atomic force microscopy.

Questions for 6 marks

1. What is the maximum relative centrifugal force applied when red blood cells are sedimented at l000 rpm in a rotor of maximum sample radius equal to l0 cm?
2. What is the principle of ion exchange chromatography and how is it used to purify proteins?
3. What is Beer-Lambert's law? What is its role in the study of macromolecules using UV-VIS spectrophotometer?
4. What is FRET? Explain its significance with a suitable example.
5. A solution of transmittance of 20%, when taken in a cell of 2.5 cm thickness. Calculate its concentration, if the molar absorption coefficient is l2,000 dm^3/mol/cm.
6. Calculate the relative centrifugal force (RCF) exerted on top and the bottom of a fixed angle rotor, dimensions for the maximum radius (r_{max}) at the bottom is 9 cm and for the minimum radius (r_{min}) is 4.8 cm, and the rotor is spinning at the l2000 rpm.
7. Diagrammatically explain components of UV/Visible spectroscopy.
8. If concentration of light absorbing substance is 5 g dm^{-3}, the molecular mass of the same substance is 410, path length of cuvette is 2 cm and transmission value is 80%. Calculate the molar extinction coefficient.
9. Diagrammatically explain the instrumental components of HPLC.
10. Explain any two methods of protein purification.
11. A pure protein (M = 67.0 kDa, vp= 0.72 cm^3 g^{-1}) is centrifuged at 10,000 rpm in an analytical centrifuge, at 5 °C and in the same buffer solution as was used in the last problem. At equilibrium, if the concentration of the protein at r = 6.5 cm is 0.8 mg/ml, what will be the protein concentration at r = 6.0 cm?
12. Describe the circular dichroism with a suitable diagram. Write a brief note about delta absorbance.
13. Explain any 2 detectors used in High performance liquid chromatography and state their advantages.

14. At 20ºC, human serum allumin has a diffusion coefficient of $6.l \times l0^{-7}$ cm^2/sec, and a sedimentation coefficient of 4.65. The density of water at 20ºC is 0.998. Calculate the MW of allumin, assuming a specific volume of 0.74 at 20ºC.
15. Justify: Iso electric focussing is ideal for separation of amphoteric substances.
16. Describe the components of IR Spectroscope.
17. Diagrammatically represent CD instrument and explain its principle.
18. Explain the term Retention factor and selectivity factor in chromatography.
19. Describe the components of SpectroFluorimeter.
20. Write a short note on Ion Exchange chromatography.
21. Explain with suitable example, how fractionation is carried out in differential centrifugation.
22. A protocol calls for centrifugation at 6000 x g. What r.p.m. should be used with an SS-34 rotor (maximum radius of l0.7 cm) to attain this g force?
23. Diagrammatically explain components of Fluorescence spectroscopy.
24. If concentration of light absorbing substance is 6 g dm^{-3}, the molecular mass of the same substance is 510, path length of cuvette is 2 cm and transmission value is 70%. Calculate the molar extinction coefficient.
25. Give the differences between Rate Zonal and Isopycnic centrifugation.
26. Write a short note on Infrared spectroscopy.
27. Differentiate between native PAGE and SDS-PAGE.
28. Predict the order of elution when a mixture containing the following compound is passed through a column containing gel that excludes all protein of MW 200000 and higher.
29. Cytochrome C (MW = 13000), Tryptophan synthase (MW = 117000), hexokinase (MW = 96000), ATP sulfurase (MW = 440000), glucose oxidase (MW = 154000) and xanthine oxidase (MW = 300000).
30. Write a short note on FRET.
31. Calculate absorption and transmission of 5×10^{-5} M ATP solution in 1 cm cuvette with molar extinction coefficient of $1.54 \times 10^4 dm^3/mol/cm$.
32. Explain in brief isocratic and gradient elution used in HPLC.
33. Diagrammatically represent analytical ultracentrifuge and explain its working.
34. Explain the working of IR spectrophotometer with the help of diagram.
35. Diagrammatically explain components of Electron microscopy.
36. Diagrammatically explain components ofImmunoelectron Microscopy.
37. Diagrammatically explain components of Confocal Microscopy.
38. Diagrammatically explain components of Atomic force microscopy.
39. Diagrammatically explain components of Phase contrast microscopy.

Questions for 12marks

1. A mixture of lysine, glycine, alanine, isoleucine and glutamic acid are separated by ionicexchange chromatography. What is the order of elution of these amino acids if you usegradient buffer system from pH 10 to pH 2: a) with a cation exchange resin? b) with an anionexchange resin?

2. Protein A, B, C and D having a MW of 16500, 35400, 88000and 120000 move 6.9 cm,5.8 cm, 4.6 and 1.3 cm respectively, when electrophoresed through a gel. What is molecular weight of protein E which moves 2.8 cm in same gel?

3. An aliquot of a solution containing a light absorbing substance at a concentration of

5 gm dm^{-3} was placed in a 2 cm light path cuvette. The cuvette was placed in a spectrophotometer and a beam of light of wavelength λ was passed through the cuvette containing solution. A transmission value of 80% was recorded. What is the absorbance of solution? What is molar extinction coefficient of solution?

4. Explain the principle of Electrophoresis technique used for separation of biomolecules with the example of NATIVE PAGE. Explain also, how NATIVE PAGE is different than the SDS PAGE?

5. Explain in brief following chromatographic performance parameters,

    a. Partition coefficient
    b. Selectivity
    c. Resolution
    d. Column efficiency.