

Sl. No. of Ques. Paper : 1318

F-7

Unique Paper Code : 2491504

Name of Paper : Techniques for Cellular and Molecular Biology Research

Name of Course : B.Sc. (H) Biochemistry

Semester : V

Duration : 3 hours

Maximum Marks : 75

(Write your Roll No. on the top immediately on receipt of this question paper.)

Attempt five questions in all. Question No. 1 is compulsory.

1. (A) State whether the following statements are true or false. Justify your answer in both the cases.

(i) A denser particle will have a higher sedimentation coefficient.

(ii) DNA can be visualized on the agarose gel using ethidium bromide staining.

(iii) In fluorimetry emitted light has a shorter wavelength than incident light.

(iv) Swinging bucket rotors are the rotors of choice for density gradient centrifugation.

(v) HPLC gives better resolution than LPLC.

(vi) TLC is more sensitive than paper chromatography.

(vii) Ammonium sulphate is commonly used for salting out.

(viii) A spacer arm is interposed between the ligand and the matrix in affinity chromatography. 2×8=16

(B) Define the following:

(i) Void volume

(ii) Retention time

(iii) Quantum yield. 1×3=3

2. (a) Discuss the principle of SDS-PAGE. How does the stacking and separating gel differ in terms of composition and function? Is this electrophoresis done horizontally or vertically and why?

(b) Describe any *three* different types of detectors to be used in gas liquid chromatography.

- (c) An enzyme examined by means of gel filtration chromatography in aqueous buffer at pH 7.0 had an approximate molecular weight of 160 KD. When same sample was examined by SDS PAGE, a single band of molecular weight of 40 KD was found. Explain why.
- (d) How is the void volume determined in gel filtration chromatography? 5,4,3,2
3. (a) How does organic solvent help in differential protein precipitation?
- (b) What are the *two* methods of polymerization of acrylamide monomers in PAGE? Explain.
- (c) Enlist the properties of a good matrix material. Describe *five* commonly used matrices in LPLC along with their chemical composition.
- (d) Name the *two* types of monochromators used in spectrophotometer. Discuss their limitations and advantages. 3,2,5,4
4. (a) Differentiate between hyperchromicity and hypochromicity with respect to DNA denaturation.
- (b) What is sedimentation velocity? Discuss the factors affecting sedimentation velocity of a particle.
- (c) Transmittance of a solution containing 10^{-5} M ATP is 0.75 (75%) at 260 nm in a 1 cm pathlength. Calculate its absorbance.
- (d) What are the criteria of selection of a suitable buffer to maintain the pH in ion exchange chromatography? 4,4,3,3
5. Differentiate between the following:
- (i) Western and Southern Blotting
- (ii) Cation and Anion Exchangers
- (iii) Fluor and Chromophore
- (iv) Lyophilisation and Dialysis. 4,4,3,3
6. Write short notes on the following:
- (i) Protein staining
- (ii) Paper electrophoresis
- (iii) Gel filtration chromatography
- (iv) Density gradient centrifugation. 3.5×4=14

7. (a) Give schematic diagram of a spectrofluorimeter. Explain extrinsic and intrinsic fluors with examples. How does quenching affect fluorescence?
(b) Explain Beer Lambert's law. State its limitations.
(c) Which form of affinity chromatography involves the use of metal ions? Discuss its methodology and applications. 6,4,4
8. Write the roles of the following:
- (i) Cesium chloride in density gradient centrifugation
 - (ii) Vacuum pump in ultracentrifuge
 - (iii) Silanization in GLC
 - (iv) Sucrose in sample buffer
 - (v) Guard columns in HPLC
 - (vi) Quartz cuvette in DNA estimation
 - (vii) Ammonium per sulphate (APS) in PAGE.

2×7=14