Unit-I

Absorption and Emission of Radiation

Write the answers of the following questions. (Each question carries one mark.)

1. Explain the term electromagnetic.
   The radiation which is made up of electrical and magnetic wave which are in phase and perpendicular to each other and to the direction of propagation.
2. Define wavelength and give its unit.
   The distance along the direction of propagation for one complete cycle is known as wavelength (λ). Wavelength may be expressed in centimeters (cm), micrometers (μm), nanometers (nm) or Angstrom units (Å).
3. Give the principle of Lambert-Beer's Law.
   Lambert-Beer's Law states that the amount of light absorbed is proportional to the concentration of absorbing substance and to the thickness of absorbing material or pathlength.
   $A = \varepsilon cl$
4. Define the term transmittance.
   Transmittance is the fraction of incident light which is transmitted. It is the ratio of light passing through the light incident on the specimen.
   $T = I/I_0$.
5. Explain the term molar extinction coefficient.
   The term molar extinction coefficient or molar absorption coefficient is a measurement of how strongly a chemical species or substance absorbs light at a particular wavelength.
   $\varepsilon = A/cl$.
6. Explain the quantum theory.
   The quantum theory of light states that light travels in bundles of energy known as photon.
7. Define chromophore.
   The chromophore is defined as any isolated covalently bonded group that shows a characteristic absorption in the ultraviolet or the visible region.
8. What is the radiant energy source of UV and visible radiation used in the UV-Visible spectroscopy?
   Radiant energy source of UV radiation- hydrogen lamp or deuterium lamp
   Radiant energy source of visible radiation- tungsten filament lamp
9. Give the function of monochromator.
   A monochromator resolves polychromatic radiation into its individual wavelengths and isolates these wavelengths into very narrow bands.
10. Explain the term nuclear resonance.
    The resonance absorption of a gamma ray by a nucleus identical to the nucleus that emitted the gamma ray. In order to change from low energy to higher energy state, nuclei must absorb the appropriate quantum of energy. Under the influence of magnetic field such nuclei absorb radiation in the radio wave region of the electromagnetic spectrum. This phenomenon is known as nuclear resonance or nuclear magnetic resonance.
11. Which events produce the electromagnetic radiation?

Electromagnetic radiation is produced by events at the molecular, atomic or nuclear levels. It includes oscillations of nuclei and electrons in electrical or magnetic fields, molecular bending and vibration, excitation of orbital electrons, ejection of an inner orbital electrons and rearrangement of other electrons, nuclear breakup.

12. Write the essential components of spectrophotometer.
    1) A stable and cheap radiant energy source or radiation source
    2) A monochromator
    3) Cuvettes to hold the sample along with sample holder
    4) A photosensitive detector
    5) Amplifier and recorder.

Fill in the blanks. (Each question carries one mark.)

1. _____ is the number of complete cycles occurring per centimeter. (wave number)
2. _____ are the groups which by themselves do not act as chromophores but whose presence brings about a shift of absorption band towards longer wavelength. (auxochromes)
3. The range of wavelength for visible region is between _____. (400-780 nm)
4. The wavelength at which compound absorbs the most is known as _____. (wavelength maxima λmax)
5. Absorption shares the _____ relationship with sample concentration. (linear or direct)
6. A beam of radiation from electric bulb consists of several wavelengths called _____. (polychromatic light)
7. As per the molecular vibration theory, asymmetric molecule which contains n number of atoms will have _____ modes of fundamental transitions. (3n+6)
8. The compound which absorbs in the ultraviolet range might emit radiation in the visible range, this shift towards a longer wavelength is known as _____. (stroke's shift)
9. Frequency of the radiation is usually expressed in the unit of _____. (Hertz or cycles per second)
10. Introduction of a fluorescent group into the molecule at the desired location is known as _____ fluorescence. (external fluorescence)
11. _____ is used as source of light in ESR spectroscopy. (klystron source)

Multiple choice questions. (Each question carries one mark.)

1. Speed of light is _____. (a)
    a) 3.0×10^8 m/s
    b) 3.0×10^8 cm/s
    c) 3.8×10^8 m/s
    d) 3.0×10^6 m/s
2. Light is supposed to have which type of characteristic? (b)
    a) Beam and light
    b) corpuscular and waveform
    c) Both a and b
    d) None of the above
3. 1 Å unit = _____ (b)

a) 10^{-8} m
b) 10^{-8} cm
c) 10^{-6} m
d) 10^{8} cm

4. Relationship between transmittance and sample concentration is _____. (b)
   a) Linear
   b) Non-linear
   c) Perpendicular
   d) Parallel
5. _____ is/are most commonly used sources of infrared radiation. (d)
   a) Nernst glower
   b) Globar
   c) Mercury lamp
   d) Both a and b
6. IR region is associated with _____ type of transition of molecules. (a)
   a) Vibrational
   b) Rotational
   c) electronic
   d) All of the above
7. Vibrational frequency should _____ with strength of the bond. (c)
   a) Decreases
   b) Remains same
   c) increases
   d) transfers
8. What is the wavelength range of UV spectrum? (b)
   a) 100 nm to 500 nm
   b) 200 nm to 800 nm
   c) 300 nm to 700 nm
   d) 400 nm to 1500 nm
9. The photon of wavelength 400 nm corresponds to _____ wave number. (b)
   a) 20000 cm^{-1}
   b) 25000 cm^{-1}
   c) 40000 cm^{-1}
   d) None of the above
10. As per Beer's law, optical density is _____ to the sample concentration if the path length is constant. (a)
    a) Directly proportional
    b) Inversely proportional
    c) not proportional
    d) same as

Write the answers of the following questions. (Each question carries 8 marks.)

1. Describe the principle, instrumentation and working of UV-visible spectrophotometer.
2. Explain the instrumentation of Infrared spectroscopy.
3. What is absorption maximum? Does the structural character and the position of absorption maximum depends only upon the structure of the compound? Discuss.
4. Describe briefly the theory of NMR spectroscopy with applications. What information can be obtained from the nmr absorption peaks?
5. Explain the working of fluorescence spectroscopy. Add a note on its applications.
6. Describe in detail ESR spectroscopy with applications.

Write the answers of the following questions. (Each question carries 4 marks.)

1. Explain the laws of absorption in detail along with deviations.
2. Calculate the optical density or absorbance of 0.2 moles of adenine in 4 mL volume. Path length is 1 cm and absorption coefficient is 13.3.

3. Discuss various types of electronic transitions which takes place when a given substance absorbs light energy.
4. Describe in brief the types of detection devices or detectors used in spectrophotometer.
5. Add a note on applications of UV-visible spectroscopy.
6. Discuss the structural factors which give rise to fluorescence. Why are fluorescence spectra band spectra?
7. What is meant by chemical shift? Describe briefly about shielding and deshielding.
8. Describe the principle and working of spectroflurometer.
9. Explain in detail the instrumentation of emission flame photometry.
10. Discuss the applications of NMR and ESR.

Unit-II

Isotopic tracer techniques in Biology

Write the answers of the following questions. (Each question carries one mark.)

1. Write the names of the natural radioactive elements.
   Thorium, Uranium, Radium.
2. Define atomic mass number.
   The atomic mass number is the total number of protons and neutrons in the atomic nucleus. It is denoted by A.
3. Explain the term radioactivity?
   Radioactivity is act of emitting radiation spontaneously. It is the process by which an unstable atomic nucleus loses energy by radiation. A material containing unstable nuclei is radioactive.
4. Define the term disintegration constant.
   For a particular isotope, the proportion of nuclei that decay in a given time is a constant known as disintegration constant.
5. Calculate the half-life of ^{131}I. The decay constant for ^{131}I is 0.0866/ day.
   8.00231 day.
6. Define phosphorescence and fluorescence.
   Phosphorescence is a type of photoluminescence, when exposed to light of a shorter wavelength, a phosphorescent substance will glow, absorbing the light and reemitting it at a longer wavelength. The phosphorescent substance does not immediately reemit radiation it absorbs. Fluorescence is the phenomenon where a molecule, after absorbing radiations, emits radiation of a longer wavelength.
7. What is the use of quenchers in Geiger Muller counter?
   Geiger Muller tube sometimes might give continuous discharge due to high voltage and formation of ionization avalanche. The quenching agents or quenchers prevent continuous ionization. If continuous discharge is not prevented, the detector will not respond to any other particle except the very first which enters the tube.
8. How to determine the counting efficiency?
   Counting efficiency = $\frac{\text{counts per minute of the radioactive standard}}{\text{Disintegration per minute of the radioactive standard}} \times 100$
9. Why fluors are used in the scintillation counting.
   When electron descends to the ground state from higher level, it emits an electromagnetic radiation. So certain compounds like fluors, fluoresce when excited. Fluors are used to emit radiation of longer wavelength which are easily detected by the detector as compared to the radiation emitted by actual substance or molecule who emits shorter wavelength which might be too short to detect.
10. Explain positron emission.
    The positron is positively charged beta (β) particle. During positron emission, a proton is converted to neutron resulting in the ejection of positron. Positron emission results in loss of proton and gain of neutron.
11. Give an ideal example of alpha emission.

Alpha particles are heavier than any other particles. Alpha emission results in a decrease in atomic number of two and decrease in mass number of four along with helium atom.
Uranium (A= 238, Z= 92) when undergoes alpha emission it gives Thorium (A= 234, Z= 90) and helium atom.

12. What is the half-life of the element?
For a given isotope, time required for the original activity to fall by a half is half-life of that element.

Fill in the blanks. (Each question carries one mark.)

1. The ratio of _____ is important in determining whether an element is radioactive or not. (neutrons to protons)
2. In negatron emission, the atomic number will _____ by 1. (increase)
3. Positron emission results in loss of _____ and gain of _____. (proton, neutron)
4. The SI unit of radioactivity is _____. (Becquerel)
5. In nuclear reactor, there is bombardment of target nuclei with _____. (neutral species like neutrons)
6. The phenomenon of fluorescence due to excitation by radioactivity is known as _____. (scintillation)
7. Solid scintillation counter is particularly used for measurement of _____ emitters. (γ)
8. _____ and _____ are types of beta decay. (positron and negatron)
9. Uranium-238 shows _____ type of decay. (alpha emission)
10. The decay constant is represented by the symbol _____. (λ)

Multiple choice questions. (Each question carries one mark.)

1. Phenomenon of radioactivity was accidently discovered by French scientist _____. (a)
   c) Henry Becquerel c) Madame Curie
   d) Thomas Edison d) Soddy
2. _____ are the elements which have different atomic number but have same mass number. (c)
   c) Isotopes c) Isobars
   d) Nucleus d) Protons
3. In negatron emission, _____ is converted to _____ resulting in the ejection of negatron. (b)
   c) Neutron, neutron c) proton, neutron
   d) Neutron, proton d) neutron, electron
4. Alpha emission results in decrease in atomic number by _____ and decrease in mass number by _____. (c)
   c) 1, 2 c) 2, 4
   d) 4, 2 d) 0, 4
5. Interaction of alpha particle with matter will result predominantly in _____. (a)

c) Ionization
d) Excitation
c) None of the above
d) Both a and b

6. Liquid scintillation counting is extremely useful for quantitating _____. (b)
   c) Gamma emitters
   d) Soft β emitters
   c) α emitters
   d) β emitters
7. Half-life of carbon-14 isotope is _____. (b)
   c) 5500 years
   d) 5000 years
   c) 5 years
   d) 50000 years
8. In which decay the Helium nucleus emitted? (a)
   c) Alpha decay
   d) Gamma rays
   c) Negatron emission
   d) Positron emission
9. Spontaneous emission of highly energetic radiation having high penetrating power is known as_____. (c)
   c) Alpha radiation
   d) Radioactivity
   c) beta radiation
   d) gamma radiation
10. An alpha particle is same as _____. (a)
    c) A helium nucleus
    d) A hydrogen nucleus
    c) a proton
    d) a positron
11. Radioactivity is the characteristic of which of the following? (a)
    a) Nucleus
    b) Electron
    c) proton
    d) neutron

Write the answers of the following questions. (Each question carries 8 marks.)

1. Describe the kinetics of half-life period of radioactive disintegration.
2. Describe in detail the principle and working of Geiger Muller Counter.
3. Explain in detail the liquid scintillation counter.
4. Describe the photographic methods of measurement of radioactivity.
5. Discuss the applications of radioisotopes in biology.

Write the answers of the following questions. (Each question carries 4 marks.)

1. Describe various types of radioactive disintegration.
2. Calculate the decay constant of ^{32}P. The half-life of ^{32}P is 14 days.
3. Discuss the process of autoradiography.
4. Describe in brief the solid scintillation counting.
5. What are the advantages and disadvantages of scintillation counting?
6. How the phenomenon of phosphorescence which is used in scintillation counting is different from fluorescence?

Unit-III

Chromatography

Write the answers of the following questions. (Each question carries one mark.)

1. What is chromatography?
   Chromatography is a laboratory technique for separation of a mixture into its components. In this two mutually immiscible phases are brought into contact with each other. It is based on differential migration of the various components of a mixture through two immiscible phases.
2. Define the term partition coefficient.
   Partition coefficient also known as distribution coefficient is a term used to describe the way in which a given compound distributes or partitions itself between two immiscible phases. It is expressed as K. it is the ratio of concentration of compound in a mixture of two immiscible solvents at equilibrium.
3. Name some chromatographic technique.
   Adsorption chromatography
   Partition chromatography
   Paper chromatography
   Thin layer chromatography
   Affinity chromatography
   Column chromatography
   Gel filtration chromatography
   Gas chromatography
   Ion exchange chromatography
4. Define the term Rf value.
   The Rf value of a compound is equal to the distance travelled by the compound divided by the distance traveled by the solvent front from the baseline or origin. The retardation factor is used to compare and help identify compunds.
5. Enlist the various factors that affect the Rf value of a compound.
   Nature of the solvent, sample size, temperature, absorbent, nature of the compound
6. What are the fundamental features of the compound used as a developer?
   It should be volatile.
   It should impart color to the different spots.
   It should not react with various compounds which are being separated.
7. Define eluant and elution.
   Eluant is the carrier portion of the mobile phase. It moves the analyte through the chromatograph.
8. Name some common adsorbent used in adsorption chromatography.
   Powdered cellulose, starch, sucrose, calcium carbonate, magnesia, silica gel, and alumina.
9. Define ion exchange chromatography.

Reversible exchange of ions in solution with ions electrostatically bound to the inert support medium is ion exchange chromatography. It separates ions and polar molecules based on their affinity to the ion exchanger.

10. Explain the term affinity chromatography.
    Affinity chromatography is a separation method based on a specific binding interaction between an immobilized ligand and its binding partner. Examples include antibody/antigen, enzyme/substrate, enzyme/inhibitor interactions.
11. Define adsorbent.
    An adsorbent is a solid substance used to collect solute molecules from a liquid or gas.
12. Chromatography cannot be used to separate delicate products (True or False).
    False

Fill in the blanks. (Each question carries one mark.)

1. The chromatography was discovered by Russian scientist _____. (Michael Tswett)
2. The pattern on the paper in paper chromatography is called _____. (chromatogram)
3. By the mean of _____ action, the liquid rise through a filter paper in paper chromatography. (capillary)
4. Chromatography with solid stationary phase is called _____. (adsorption chromatography)
5. Thin layer chromatography is _____ chromatography. (adsorption chromatography)
6. In thin layer chromatography, the stationary phase is made up of _____ and the mobile phase is made up of _____. (solid, liquid)
7. When the single solvent is used as an eluent during development, the process is known as _____ elution.
8. The pattern on the paper in paper chromatography is called _____. (chromatogram)
9. _____ is used as a spraying reagent in paper chromatography. (ninhydrin spray or solution)
10. _____ is used as a carrier gas in gas chromatography. (inert gas or hydrogen)

Multiple choice questions. (Each question carries one mark.)

1. The compound which interacts more with the mobile phase and least with stationary phase _____. (a)
   a) Migrates Fast
   b) Migrates Very slow
   c) remains steady
   d) None of the above
2. On which factors does the Rf value of a compound depend? (d)
   a) Nature of the compound
   b) Temperature
   c) nature of the solvent
   d) All of the above
3. Paper chromatography is a separatory technique that is used to separate _____ (b)
   a) Simple mixtures
   b) Complex mixtures
   c) viscous mixtures
   d) metals

4. The size of the spot in paper chromatography should be _____. (a)
   a) 1 – 2 mm
   b) 6 – 8 mm
   c) 2 – 5 mm
   d) 8 – 9 mm
5. Which force is involved in the chromatography? (d)
   a) London force
   b) Hydrogen bonding
   c) Electric static force
   d) All of the above
6. Ion exchanged chromatography is based on _____. (c)
   a) Electrostatic attraction
   b) Adsorption chromatography
   c) electrical mobility of ion species
   d) partition chromatography
7. The columns used in column chromatography are usually made up of _____. (d)
   a) Glass
   b) Polyacrylate plastic
   c) Both a and b
   d) either a or b
8. Which of the following columns are not used in liquid or high performance liquid chromatography? (d)
   a) Analytical column
   b) Guard column
   c) separation column
   d) capillary column
9. The mobile phase used in the chromatography can be made up of _____. (b)
   a) Solid or liquid
   b) Liquid or gas
   c) gas only
   d) liquid only
10. Chromatography with solid stationary phase is called _____. (d)
   a) Circle chromatography
   b) Liquid chromatography
   c) solid chromatography
   d) adsorption chromatography

Write the answers of the following questions. (Each question carries 8 marks.)

1. Explain in detail affinity chromatography.
2. Describe briefly the working of gel filtration chromatography.
3. Explain in brief paper chromatography.
4. Explain column chromatography.
5. Describe in brief the ion exchange chromatography along with applications.
6. Explain the instrumentation and applications of high performance liquid chromatography.
7. Elaborate in detail molecular sieving chromatography.

Write the answers of the following questions. (Each question carries 4 marks.)

1. Write a note on instrumentation of gas chromatography.
2. Enlist the applications of affinity chromatography.
3. Describe the sampling, detection and analysis of the analytes in thin layer chromatography.
4. Comment on the various types of gels used for gel filtration chromatography.
5. How to determine molecular weight by gel filtration chromatography.
6. Discuss the use of affinity chromatography in molecular biology.

Unit- IV

Electrophoresis

Write the answers of the following questions. (Each question carries one mark.)

1. Explain the term electrophoresis.
   Electrophoresis is migration of charged particles or molecules in a medium under the influence of an applied electric field. It is used as separation method.
2. Enlist commonly used buffers in electrophoresis.
   Tris acetate EDTA (TAE)
   Tris borate EDTA (TBE)
3. What is the role of SDS in SDS-PAGE?
   Protein denaturing and imparting net negative charge.
4. Enlist the factors affecting electrophoresis.
   Molecular charge, molecular shape and size, strength of the electrical field, ionic strength, viscocity, temperature of the medium.
5. Define the term adsorption.
   Adsorption is the adhesion of atoms, ions or molecules from a gas, liquid or dissolved solid to a surface. This process creates the film of the adsorbate on the surface of adsorbent.
6. What is isoelectric focusing?
   Isoelectric focusing is a technique for separating different molecules by differences in their isoelectric point within a continuous pH gradient.
7. Define isoelectric point of protein.
   Isoelectric point of protein is defined as the pH at which the net charge of a protein molecule is zero. The molecule carries no electrical charge hence does not migrate towards any electrode under the influence of electric field.
8. What agarose concentration should be used in the preparation of gel?
   The concentration of agarose in gel will depend on the sizes of DNA fragments to be separated ranging within 0.5% to 2%.
9. Give the principle of immunoelectrophoresis.
   An electrophoresis for separation of the protein antigen in semisolid media followed by an immunodiffusion against the antiserum resulting in precipitin formation. The technique exploits the specificity of reaction between an antigen and antibody.
10. Define the term electrophoretic mobility.
    It is defined as being the field strength reduced velocity of the ions in the capillary. Charge/mass ratio of the sample dictates its electrophoretic mobility.

Fill in the blanks. (Each question carries one mark.)

1. The speed of migration of ions in an electric field depends upon magnitude of _____. (charge and shape of molecules)
2. In SDS-PAGE separation is based on _____. (molecular weight)
3. Electrophoresis was developed by _____. (Tiselius)

4. The electrophoretic mobility denoted as μ is mathematically expressed as _____. (V/E, ratio of velocity of the biomolecules in the electric field and it's intensity)
5. Electrophoresis is not used for separation of _____. (lipids)
6. In SDS-PAGE of protein separation, one SDS molecule will binds to _____. (every two amino acids)
7. Proteins can be visualized directly in gels by _____. (staining them with the dye)
8. In SDS-PAGE, migration of protein is affected by _____. (size of protein)
9. The tracking dye used in SDS-PAGE will be _____. (anionic)
10. Fragments of DNA in gel electrophoresis travel away from the negative electrode, this is because DNA is _____ charged so attracted to the _____ end. (negative, positive)

Multiple choice questions. (Each question carries one mark.)

1. In electrophoresis, DNA will migrate towards _____. (b)
   a) Cathode or positive electrode
   b) Anode or positive electrode
   c) Cathode or negative electrode
   d) Anode or negative electrode
2. The most common type of gel used for DNA separation is _____. (b)
   a) Agar
   b) Agarose
   c) polyacrylamide
   d) All of the above
3. In isoelectric focusing, proteins are separated on the basis of their _____. (a)
   a) Relatively content of positively and negatively charged residue
   b) Relatively content of positively charged residue only
   c) Relatively content of negatively charged residue only
   d) size
4. _____ ratio of the sample dictates its electrophoretic mobility. (a)
   a) Charge/mass
   b) Charge/charge
   c) mass/mass
   d) None of the above
5. In electrophoresis, rate of migration of biomolecules is _____. (b)
   a) directly proportional to the Mass
   b) directly proportional to the current
   c) inversely proportional to the current
   d) directly proportional to the resistance
6. Which of the following will migrate faster? (d)
   a) Nicked circular DNA
   b) Double stranded DNA
   c) single stranded DNA
   d) Supercoiled circular DNA
7. Which of the following factors does not influence electrophoretic mobility? (c)
   a) Molecular weight
   b) Shape of the molecule
   c) stereochemistry of molecule
   d) size of the molecule
8. Which of the following cannot be used for the separation of nucleic acids? (a)
   a) SDS-PAGE
   b) Northern blotting
   c) PAGE
   d) All of the above
9. Sodium dodecyl sulfate (SDS) used in SDS-PAGE is _____. (a)
   a) An anionic detergent
   b) Non-ionic detergent
   c) cationic detergent
   d) an anion exchanger

10. Which of the following is used as tracking dye in SDS-PAGE of protein? (d)
    a) Bromophenol blue
    b) Orange G
    c) xylene cyanol
    d) All of these

Write the answers of the following questions. (Each question carries 8 marks.)

1. Take a detail insight to isoelectric focusing.
2. Describe SDS-PAGE electrophoresis method with its applications.
3. Explain in detail paper electrophoresis.
4. Elaborate the method and instrumentation of gel electrophoresis.
5. Describe immunoelectrophoresis.
6. Is it possible to separate on the basis of isoelectric point? If yes, discuss the method in detail.

Write the answers of the following questions. (Each question carries 4 marks.)

1. Add a note on moving boundary electrophoresis.
2. Discuss in detail applications of paper electrophoresis.
3. Discuss various types of gels used for gel electrophoresis.
4. Add a note on applications of gel electrophoresis in detail.
5. How to separate proteins on the basis of their molecular weight and charge.