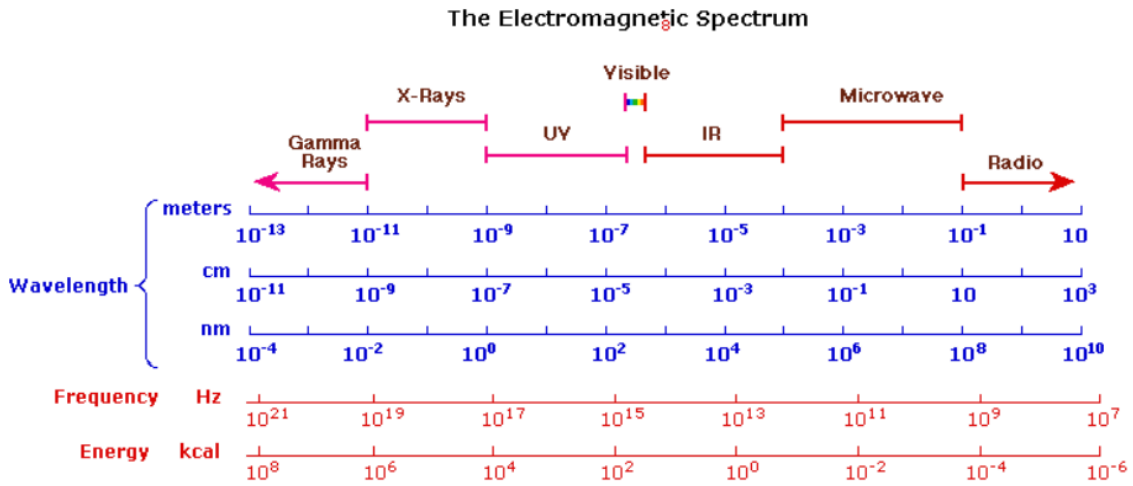
Spectroscopy



Weightage: 15 marks compulsory question

Energy is proportional to the frequency of waves.

$$f = \frac{c}{\lambda}$$

$$E = hf$$

The amount of light absorbed depends on the concentration of the solution and the length traversed by the light through the solution.

Lambert's Law: The law states that "**Equal fractions** of the incident light are absorbed by successive layers of **equal thickness of the absorbing medium"**

If a monochromatic light passes through a transparent medium, the rate of decrease in intensity with the thickness of medium is proportional to intensity of the incident light

$$-\frac{di}{I} \alpha\, dx$$

$$\ln(If / Ii) = - k.\text{length of medium}$$

$$log\frac{I\,initial}{I\,final} = \frac{k \times length\ of\ medium}{2.303}$$

Beer's law: The law states that "**Equal fractions** of the incident light are absorbed by successive layers having **equal concentration** of the absorbing medium".

$$-\frac{di}{I} \alpha\, dc$$

$$\ln(If / Ii) = - k.\text{concentration of medium}$$

$$log\frac{I\,initial}{I\,final} = \frac{k \times concentration\ of\ medium}{2.303}$$

Beer Lambert's Law: Equal fractions of the incident light are absorbed by successive layers of **equal thickness** and **equal concentration** of the absorbing medium.

$$A = log \frac{I\ initial}{I\ final} = \frac{k \times concentration\ of\ medium \times length\ of\ medium}{2.303}$$

$$\varepsilon = \frac{k}{2.303}$$

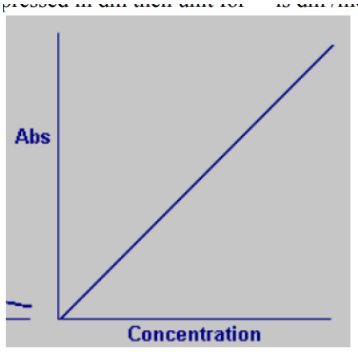
$$A = log \frac{1}{T} = log \frac{I\ initial}{I\ final} = 2 - \log \%T$$

$$T = \frac{I\ final}{I\ initial}$$

//all logs are log base 10

1. 7.5 x 10-4 M solution of an absorbing species is placed in a cell of path length 2 cm. Transmittance measured is 60 % at 470 nm. Calculate the molar absorptivity?
2. If the transmittance of the solution is 19.4 %, what will be its absorbance ?
3. The transmittance of 2 x 10-4 M solution was found to be 76.2 % in path length of 1 cm. calculate the i) absorbance ii) Molar absorptivity iii) % T for the path length of 2 cm.

Monochromatic: Light of a single wavelength



Limitations of Beer Lambert's Law

1. The linearity of Beer Lambert law is limited by chemical/instrumental factors

Chemical:

- Law isn't valid for solutions with **conc more than .01 M.** Solution: Dilute for higher concentration and adjust factor in calculation.
- **Molar extinction coefficient (epsilon) depends on the refractive index** of the solution. RI changes with solvents. Solution: do not use different solvents in our experiments.
- Interaction with solvent occurs in the form of **Hydrogen Bonding**. Absorbance is proportional to concentration. Due to HBonding, 2 molecules merge into one, thus there's a misunderstanding in the actual to intended concentration. Solution: Hydrogen bonding is unfavorable in nonpolar solvents, thus we use nonpolar solvents.
- **Scattering of light** due to particulate matter in samples. If light scatters, it wont pass through the detector. There will be a loss in reading absorbance. Solution: Filter the solution beforehand.
- **Fluorescence and phosphorescence**: It is a delayed emission of light. After fluorescence, extra light is emitted (in the form of transmittance and radiation due to excitation of electrons) which looks as if absorption is reduced but actually it's much higher. In phosphorescence, stepwise the excited electrons come down to the energy level and there is a deviation. Thus, a positive deviation in T and a negative deviation for A is observed. Solution: Use instruments like fluorescence spectrophotometer and phosphoroscence spectrophotometer.

- Shift in chemical equilibrium in terms of concentration. Solvents should not normally prefer keto-enol, or other types of **tautomerism**, lest they alter absorption readings. Thus, we avoid polar solvents. Use nonpolar like CCl4, toluene, benzene, etc.

2. Physical factors:
   - If a **non monochromatic** radiation is passed, there will be a deviation. This can be minimized by a flat part of the absorption spectrum.
   - **Stray light**: Light from surroundings other than the beam as intended. The stray light will increase readings unnecessarily. Solution: Perform experiment in a closed space to restrict entry of stray light.

Ultraviolet-visible spectrophotometer

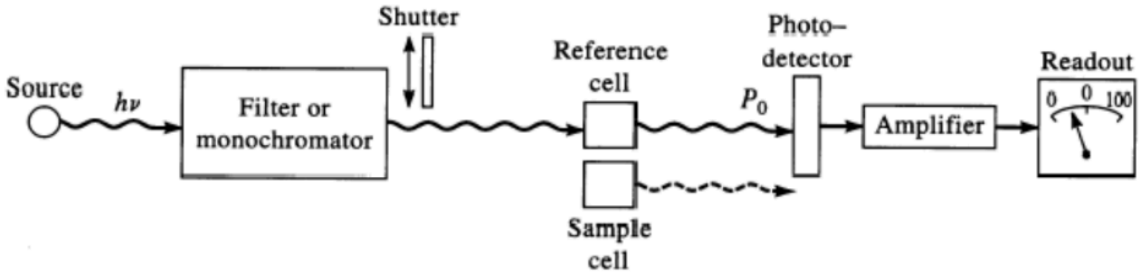
Source for lights:

1. Uv: deuterium lamp
2. visible: tungsten filament
3. both: xenon

Single Beam Spectrophotometer

If the cuvette is glass, it absorbs UV rays. To avoid this loss, we use quartz as it does not absorb uv or visible rays. It is expensive and fragile.

We can also use a transparent plastic, but it can also absorb at a certain wavelength. Reading will be lost at that wavelength.



Monochromator definition: Although prisms can be used to separate light into different wavelengths, most instruments use diffraction gratings. When light hits the grooves on a diffraction grating at an angle, it gets split into different wavelengths consistently, as long as the grooves are made uniformly.

Detectors:

1. **Photomultiplier Tubes (PMTs)**: These detect light by turning photons into electrons, which are then amplified. PMTs are very sensitive, especially at around 400 nm, and are often used in instruments that measure one wavelength at a time.
2. **Photodiodes**: Less expensive and less sensitive than PMTs, photodiodes are used in arrays to detect light at multiple wavelengths at once.
3. **Charge-Coupled Devices (CCDs)**: These are very sensitive detectors that store charges from light. They can gather data over a longer period and are becoming cheaper and more common in spectrophotometers, possibly replacing PMTs in some cases.

Cuvette:

1. Cuvettes are containers (made of plastic, glass, or quartz) used to hold liquid samples for UV-Visible spectroscopy.

Working:

In a single beam spectrophotometer, we first put solvent in the reference cell. Upon passing a beam from the filter, some light gets absorbed into the solvent, the resultant beam falls on the photo-detector. This photodetector is super sensitive, and thus to effectively convert digital to electrical signal, we pass the beam through an amplifier before passing it to the reader. Then, the beam is sent through the sample cell. The entire process is repeated. This juggle between RC and SC is done for all wavelengths.

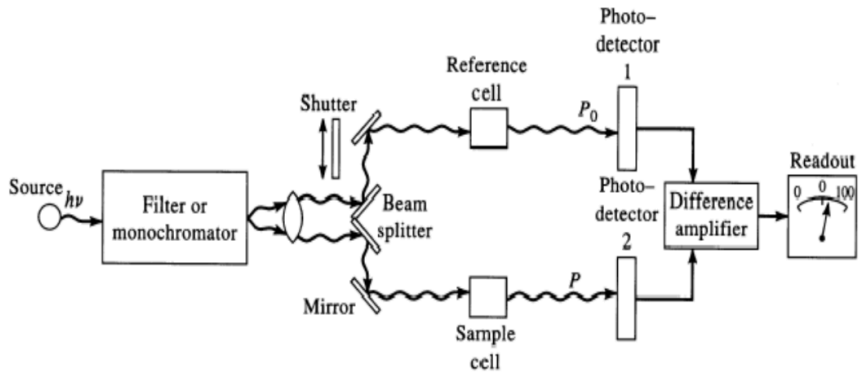
When plotted against the xy axis, the highest peak on the graph is called the wavelength of highest lambda max.

A = 2-log(%t)

Formula for conversion of absorbance to transmittance.

Single beam spectrophotometer is **inefficient due to large manual input** and thus, we use double beam spectrophotometer.

Double Beam spectrophotometer



1. Light source creates UV and visible light
2. Filter will select light of single wavelength
3. Beam splitter will split into two
4. Light will pass through Reference cell and sample cell at the same time
5. PDs detect lights from both cells
6. The difference is amplified automatically
7. The reading is sent to readout.

Advantages:

1. Compensates for Light Intensity Changes: Voltage fluctuations are balanced by splitting the beam into two identical beams, ensuring accurate readings by adjusting the blank absorbance to zero.
2. Stable Readings: The sensitivity of photodiodes doesn't affect the readings, as the zero method compensates for any variations.

3. Linear Scale: The instrument gives a linear relationship between absorbance and sample concentration.

Application of UV Visible Spectroscopy

1. Quantitative determination of analyte
2. You can identify functional groups in a molecule by measuring its **λmax**. Each functional group absorbs light at a specific **λmax**. Spectrophotometers shine light of different wavelengths, and the absorption pattern helps identify the functional groups present in the sample. Identification of inorganic and organic species. (Which compound, what components)
3. Magnitude of molar absorptivity. (if epsilon is 10k-100k, compound is organic. If less than 3k, compound is aliphatic)
4. Detector in HPLC: high performance liquid chromatography.
5. Used in semiconductor industries to measure thickness and optical properties of thin films.
6. Study absorbance of organic compounds.

Advantages of UV-Vis Spectrophotometry:

1. **Fast and simple**: Quick analysis with minimal sample preparation.
2. **Non-destructive**: Does not alter the sample.
3. **Wide application**: Used in chemical, pharmaceutical, environmental, and biological analysis.
4. **High sensitivity**: Can detect low concentrations of substances.
5. **Quantitative and qualitative**: Can determine both concentration and molecular structure.

Limitations of UV-Vis Spectrophotometry:

1. **Limited to UV-visible absorbing substances**: Only works for compounds that absorb UV or visible light.
2. **Interference from impurities**: Other substances in the sample may affect the results.
3. **Requires clear, transparent samples**: Samples must be free of turbidity or color interference.
4. **Sensitive to matrix effects**: The solvent or surrounding environment can influence measurements.
5. **Limited structural information**: Does not provide detailed molecular structure

pH Metry

Definition: pH is defined as the negative base 10 logarithm of the hydrogen ion concentration.

$$pH = -\log_{10} [H^+]$$

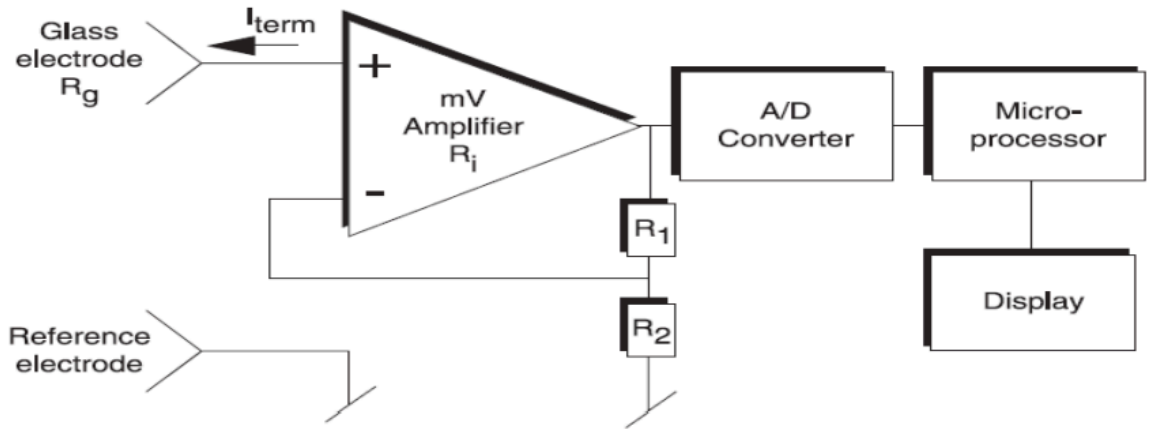
Nerst equation:

$$E = E^o - \frac{2.303\,Rt}{nF} \times log10\,[H\ +]$$

Thus,

$$E = E^o + 0.059 \times pH$$

- A reference electrode is an electrode with constant and known electrode potential: SHE or calomel



simplified principal construction of a pH meter

Measurement: A pH meter measures the pH of a solution using a pH probe, reference probe, and sometimes a temperature sensor, often combined into one electrode. When the probe touches the solution, a potential difference forms between the pH and reference probes. The meter amplifies this potential, converts it to a digital signal, and calculates the pH based on a calibration curve.

Standardization of pH Meter:

- Immerse the glass electrode in a pH 4.0 buffer and adjust the reading to 4.0 using the knob.
- Rinse the electrode, then immerse it in a pH 9.2 buffer and adjust the reading to 9.2.
- Rinse the electrode with distilled water after each use, blot it dry, and wait at least 5 minutes after turning the meter on before starting measurements.

This two-point calibration ensures accurate readings across a range of pH values.

Glass electrode:

- **Construction**: It has a glass bulb membrane with an insulating tubular body. Inside is an internal solution and an Ag/AgCl electrode connected to a pH meter via a cable. It works in contact with the solution being measured.
- **Working**: The pH meter measures the potential difference across the glass membrane. This potential depends on the hydrogen ion concentration and is given by the nernst equation.

Reference electrode

- It contains a silver wire coated with silver chloride, immersed in an electrolyte (often a gel with KCl for conductivity).
- Maintains a constant potential as long as the internal electrolyte remains stable.
- The reference potential is influenced slightly by temperature and depends on chloride ion concentration

Calomel electrode: (Reference electrode)

Construction: Take a tube. Bottom of the tube, add a mercury drop. Cover with paste of solid mercuric chloride. The aqueous phase in contact with the mercury drop and mercuric chloride (called calomel) is a saturated solution of KCl in water. The electrode is linked via a porous frit (salt bridge) to the solution in which the other electrode is immersed.

$$Hg_2Cl_2 \rightarrow Hg_2^{2+} + 2Cl^-$$

$$Cl^- (4m) \mid Hg_2Cl_2 (s) \mid Hg (l) \mid Pt$$

Working as a cathode:

½ $Hg^2Cl^2 + e^- \rightarrow Hg + Cl^-$

Anode:

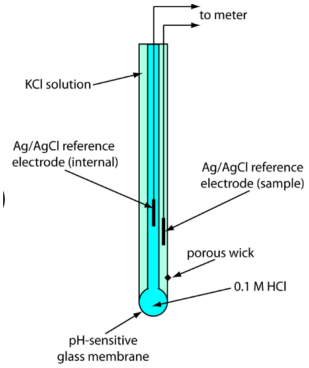
$Hg_2^{2+} + 2e^- \rightarrow 2Hg$ (written in reverse order because we consider E(reduction) only for both anode and cathode)

Thus, product is Hg_2^{2+} for nernst eqn.

Advantages of calomel electrode:

1. Simple to construct
2. Results of cell potential measurement is reproducible
3. Stable for a long time

Ion selective electrode



Definition: it is selective for a particular ion. The glass membrane is doped such that it is sensitive to one specific ion.

HCl soln dipped electrode is indicator electrode and KCl dipped electrode is reference electrode. Porous wick functions as the salt bridge.

Cell representation: Ag | AgCl | HCl | glass || probed solution | reference electrode (kcl)|AgCl|Ag

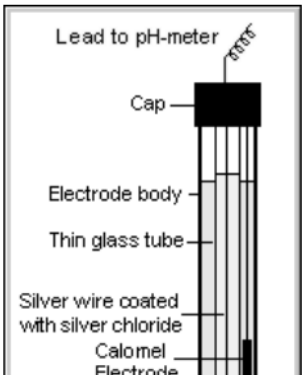
Construction: Consists of a glass membrane that separates an internal solution and silver and silver chloride from the studied solution.

pH meter measures the potential difference and the formula is already written above.

Applications:

1. To analyse foods, cosmetics
2. Comparison of indicators

Combination Electrode



Construction: The pH electrode has a thin glass bulb filled with pH 7 KCl solution. Inside, a silver wire coated with silver chloride maintains a constant reference potential.

Working: The outside surface of the glass membrane is in contact with the sample being measured and the inside surface contacts the filling solution KCl. The inner glass/filling solution potential stays constant, while the outer glass/sample interface potential changes based on the sample's H+ concentration and temperature. The difference determines the pH.

Measurement of pH

1. Two point standardisation

This implies using two buffer solutions (4,9). Take the solution in beaker. Dip the electrode in this. Measure the pH in the pH meter. Put the pH meter in standby mode after analysis. After removing the electrode, rinse with water and put in a yellow black pH buffer to avoid hampering the glass membrane. Cover it with paraffin to avoid evaporation of the buffer.

2. Three point standardization

Uses three buffer solutions (4,7,9)

Conductometry

Definition: Conductometry is a general method wherein two electrode systems are used for simultaneous measurement of all *electroactive* compounds in a solution. It is a **measurement of electrolytic conductivity** to monitor a chemical reaction's progress.

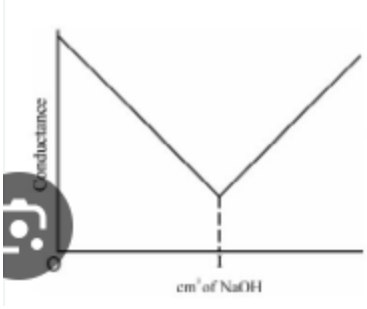
Conductometric Titration

Conductance shows how many ions are in a solution and how well they can carry electricity. If the solution has only one type of electrolyte, the conductance tells us about its concentration.

Conductometric titration measures how the solution's ability to conduct electricity changes as a reactant is added to the reaction. The **equivalence point** is where there's a sudden jump or drop in conductivity, usually because the amounts of hydrogen or hydroxyl ions change. This method works well for solutions or suspensions (like wood pulp) where regular color-changing indicators don't work.

Principle: When one electrolyte is added to another without much volume change, the conductance of the solution will change if an ionic reaction occurs. If no reaction happens, conductance simply increases. But if a reaction occurs, one ion may replace another, changing the conductance. After the equivalence point, the reaction stops, and the conductance increases due to the excess of titrant (C+D-). (Now, all replaceable ions have been replaced and adding more free ions will only increase the conductivity)

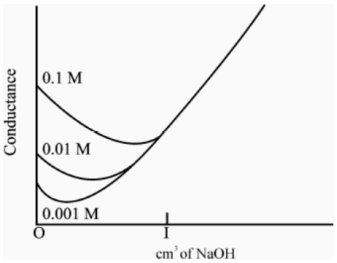
1. Strong acid and strong base (HCl vs NaOH)



*The point of minimum conductance is called the equivalence point. This is the point of neutralization.

2. Weak acid vs strong base

Conductance is decreasing at first due to dissociation of acetic acid (weak). The increase is due to the formation of sodium acetate (Strong electrolyte). Once all the NaOH has reacted with all acetic acid molecules, we add extra moles of NaOH. Upon adding any more base after point of equivalence, the graph shows a sudden increase. This sudden jump and curve's point of intersection give an equivalence point.

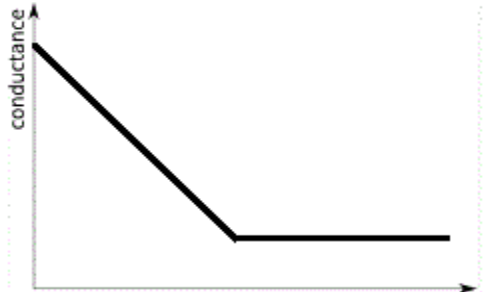


For higher molarity, the starting point is up higher on the y axis. For lower molarity, the starting point is lower on the y axis.

Higher concentration α $More\ Dissociation$

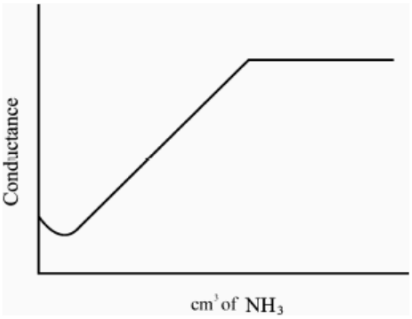
3. Strong acid weak base

The beaker has HCl already. It's already a strong electrolyte. The conductance is high. Upon adding nh4oh, the volume of hcl reduces till all of it is exhausted. At this point, only the salt in the beaker accounts for conductivity and the graph becomes flat. This is the same case when strong base is already present and weak acid is introduced.

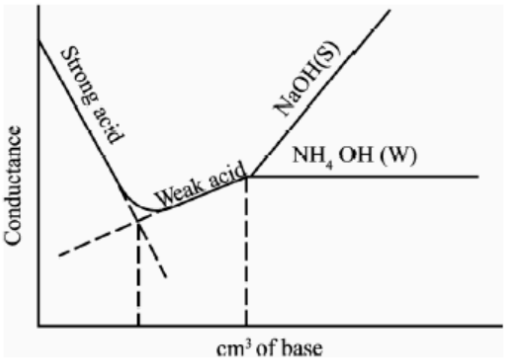


4. Weak base weak acid

After the equivalence point, conductance remains same as the weak base which is being added is feebly ionized and, therefore, is not much conducting.



Mixture



Advantages

1. **Coloured solutions** can be titrated
2. Conductometric titration works well for **dilute solutions** as well, because it relies on changes in conductance rather than the value of conductance.
3. It is **not necessary to make precise measurements** near the equivalence point, as measurements on both sides of the equivalence point are useful.
4. This method can also titrate very weak acids or bases, which may not be possible in traditional titrations.
5. Additionally, it can be used to titrate a mixture of weak and strong acids, allowing for the simultaneous determination of both.

Limitations:

1. In dilute solutions, conductometric titrations often produce obtuse curves, making it difficult to pinpoint the equivalence point accurately.
2. Additionally, the technique has limited overall accuracy because it does not allow for the addition of small increments of the titrant.

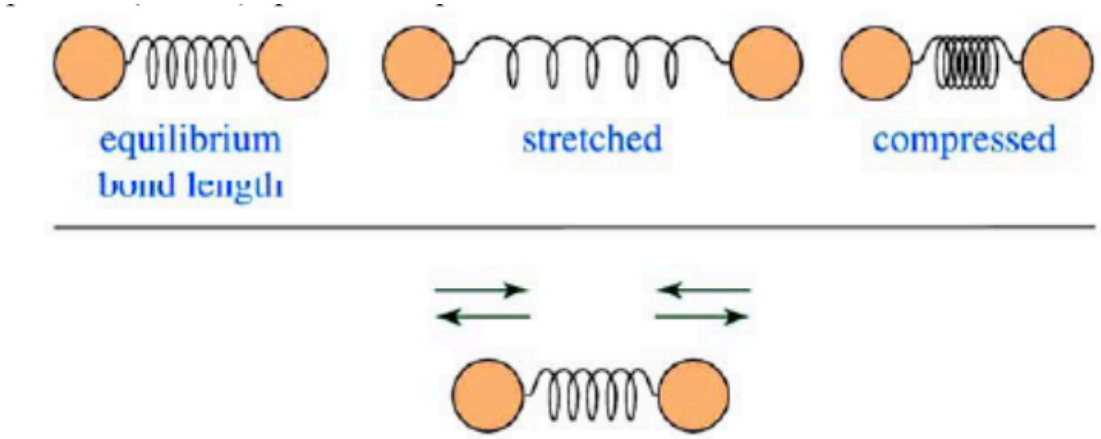
Infrared spectroscopy

Contrary to the uv spectroscopy, the IR spectroscopy offers a rich array of absorption spectra. These bands provide accurate structural information about a molecule. There are three types:

1. 15000 - 3000 : near ir
2. 4000 - 400 : mid ir
3. 200 - 10 : far ir

*unit is wave number/frequency.
Near IR → closer to visible region
The most used waves are 4000 - 670 cm^{-1}

Infrared is a highly thermal energy. It induces stronger molecular vibrations in covalent bonds. Specific bonds respond to specific frequencies in the form of absorption.



Vibrational modes

1. Stretching: the distance between two atoms increases or decreases but atoms remain in the same bond axis.
    a. Symmetric stretching: stretch is in the same direction
    b. Asymmetric stretching: stretch is in diff directions
2. Bending: the angular displacement between two atoms increases or decreases.
    a. Scissoring: angle increases or decreases in itself like a scissor motion
    b. Rocking : angles increase and decrease in the same direction simultaneously.

Different stretching and bending occurs at certain frequencies only. When an IR of some frequency is incident on the molecule, the energy is absorbed and likewise, the amplitude of vibration increases. When the molecule comes to rest, the absorbed energy is released as heat.

Which substances give a signal in the IR spectrum?

The **molecules containing polar bonds**, i.e. molecules composed of atoms of different elements like HCl, H2O, can give a signal in the IR spectrum. These compounds give **Raman spectrum peak.** Pure elements like Ar, N_2, diamond etc cannot give a signal.

Definition: The frequencies of radiation absorbed are measured by an infrared spectrometer, and the resulting plot of absorbed energy vs. frequency is called infrared spectrum of the material

Instrumentation of FTIR (Fourier Transform Infrared Spectroscopy)

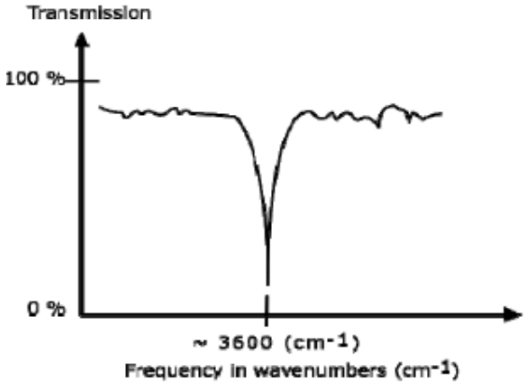
Basic components of FTIR are:

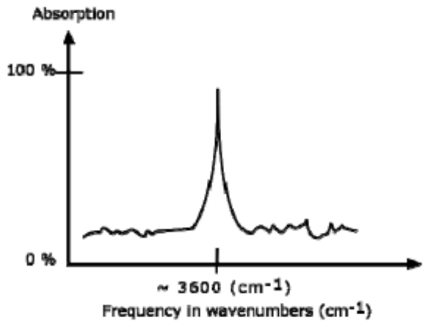
1. Source: Infrared energy is emitted from a *glowing black body source*. This beam is passed through an aperture. This aperture controls the amount of energy passed to the sample.
2. Interferometer: Here, the spectral encoding takes place. The resulting interferogram signal exits the interferometer.
3. Gas samples can be directly analysed. Liquid can also be used directly but in diluted form in NaCl plates. Solid compounds can be mixed with KBr and formed a pallet and used.
4. Detector detects the sample and gives a reading. liquid nitrogen cooled MCT (Mercury−Cadmium−Telluride) detector.
5. Signal and DP: Signal is digitalised and sent to the computer for further processing and manipulation.

The IR spectrum is a graph where:

- The **x-axis** shows the **frequencies** in **wavenumbers** (cm^{-1}). This can be for either absorption or transmission.
- The **y-axis** shows the **intensity** of transmission or absorption, often in percentage.

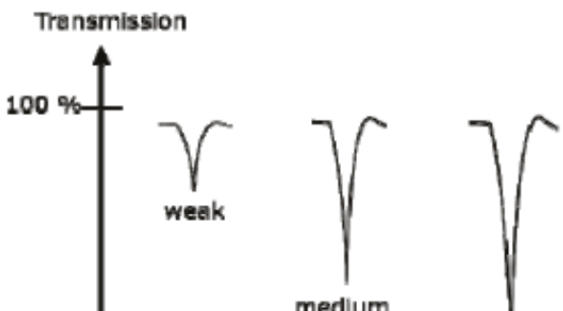
This plot represents how much infrared radiation is absorbed or transmitted by the sample at different frequencies. **Graphs:**





The graph above shows a spectrum in **absorption** mode.

Classification of IR bands

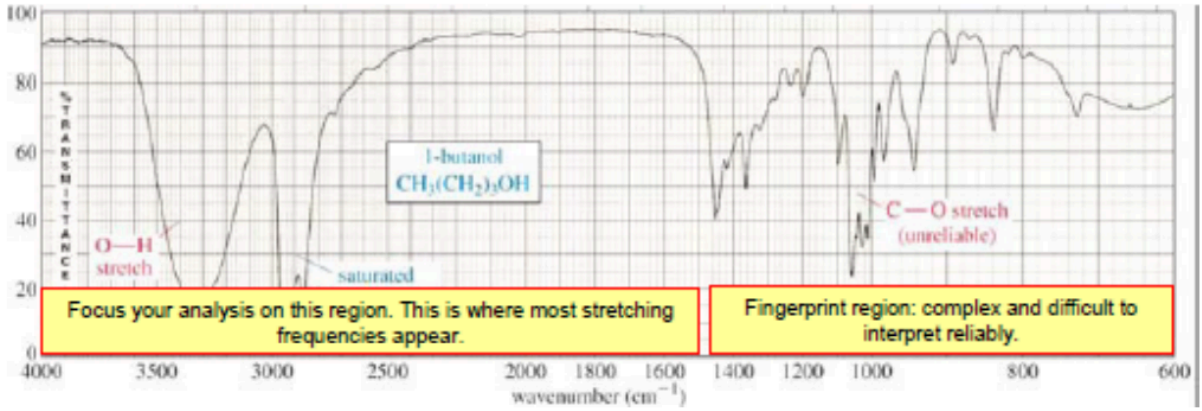


A strong band covers most of the y-axis. A medium band falls to about half of the y-axis, and a weak band falls to about one third or less of the y axis

1. 0% transmittance: strong
2. 50%transmittance : medium
3. 75% transmittance: weak
4. Broad peak: This peak shows up when there's a hydrogen bond involved.
5. Narrow peak: Most bands are narrow and sharp

Fingerprint Region

The 600 - 1400 cm-1 range is called the fingerprint region.



/// Characteristic IR Absorption Frequencies of Organic Functional Groups do from pdf

NMR

The position of proton in the spectrum helps us to know the type of proton viz. aromatic, aliphatic, alicyclic, aldehydic etc.
Definition: Chemical shift is a measure of how much the resonance frequency of a proton (hydrogen atom) in a molecule differs from a reference proton (usually TMS) in NMR spectroscopy.

Chemical shift $\delta=(Hs-Htms) / Ho \times 10^6$ ppm

Hs = resonance frequency of the sample proton.
Htms = resonance frequency of the TMS reference proton.
Ho = operating frequency of the NMR instrument.

$$\delta = \frac{\Delta v \times 10^6}{Oscillator\ frequency\ (cps)}$$ del v = change in frequency

Following factors are responsible for influencing its value:

- Specific solvent,
- Bulk diamagnetic susceptibility effect,
- Temperature (only when change in temperature causes changes in some type of association equilibrium or changes in amplitude of torsional vibrations),
- Electron density,
- Inductive effect,

- Van der Waal deshielding, and
- Hydrogen bonding.

Definition: Shielding is when the electron cloud around a proton creates a small magnetic field that opposes the external magnetic field applied in NMR. This reduces the effect of the external field on the proton, making it "shielded" and causing it to absorb at a lower frequency.
Definition: **Integration** in NMR tells you how many protons are responsible for each signal. The bigger the peak, the more protons there are. It gives a **ratio** of protons, not the exact number.

Splitting of Signals (Spin-Spin Coupling) in NMR:

- NMR signals don't always appear as simple, single peaks; they often split into multiple peaks (e.g., **doublets** with 2 peaks, **triplets** with 3 peaks, **quartets** with 4 peaks).
- The distance between these split peaks is called the **coupling constant (J)**, which gives information about the interaction between nearby protons.

How it works:

- Protons have a **spin** that can either be aligned parallel or antiparallel to the applied magnetic field.
- If two protons are close enough, their spins can affect each other, leading to different energy levels for the proton's absorption. This is called **spin-spin coupling**.
- The result is that one proton's signal gets split into multiple peaks based on how its spin interacts with nearby protons.

Reciprocal Splitting:

- If proton **Ha** splits proton **Hb**, then **Hb** will also split **Ha**, and vice versa. This helps identify the number of neighboring protons a specific proton has, providing more structural information about the molecule.

Applications

1. In NMR, the **area of a peak** tells you how many protons are responsible for it. By using a known reference (internal standard), you can calculate the concentration of a substance in your sample. This helps track how much of a compound is present or formed in a reaction.

Qualitative Analysis in NMR involves:

1. **Chemical shift values**: Identifying hydrogen groups (CH_3, OH, etc.) by their specific shifts.
2. **Functional groups**: Recognizing unique shifts from groups like OH, NH, or C=O.
3. **Relative position**: Determining the arrangement of groups based on peak positions.
4. **Number of nuclei**: Using peak size to find how many protons are in each group.