BP605T · B.Pharm Semester VI · Theory Notes

Pharmaceutical Analysis

Unit II — Spectroscopy (UV-Visible & IR)

Unit II (10 hours)

Pills & Python Notes

From-zero teaching standard: every technical term is defined before use.

Syllabus coverage: Unit II (10 hours).

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Unit overview. This unit teaches how matter interacts with light (electromagnetic radiation) and how that interaction is used to identify and measure drugs. Two chapters:

- **Chapter 4 — Fundamentals of Electromagnetic Radiation and UV-Visible Spectroscopy:** wave properties, the EM spectrum, Beer–Lambert law (full derivation), electronic transitions, chromophores/auxochromes, instrumentation and single/multi-component analysis.
- **Chapter 5 — IR Spectroscopy and FTIR:** molecular vibrations, dispersive IR and FTIR instrumentation, sample handling, and pharmaceutical applications.

CHAPTER 4 — FUNDAMENTALS OF ELECTROMAGNETIC RADIATION AND UV-VISIBLE SPECTROSCOPY

Learning Objectives

After studying this chapter, the student will be able to:

1 Define spectroscopy and explain how electromagnetic radiation behaves as both a wave and a particle.

2 List the regions of the electromagnetic spectrum in order of increasing wavelength and decreasing energy, and name the spectroscopic technique used in each region.

3 Describe the interaction of radiation with matter: absorption, emission and scattering.

4 State and derive Beer's law and Lambert's law from the basic differential relation $dI = -k \cdot I \cdot dl$, and combine them into $A = \varepsilon bc$.

5 Explain the transmittance–absorbance relationship ($A = 2 - \log T$) with derivation.

6 List chemical, instrumental and fundamental deviations from Beer's law with examples.

7 Arrange electronic transitions ($\sigma \rightarrow \sigma^*$, $n \rightarrow \sigma^*$, $\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$) in order of energy and identify which are useful in UV-Visible analysis.

8 Define chromophores and auxochromes, classify K, R, B, E bands, and explain bathochromic, hypsochromic, hyperchromic and hypochromic shifts.

9 Explain the effect of solvent on absorption spectra (polar vs non-polar on $n \rightarrow \pi^*$ *and* $\pi \rightarrow \pi^*$ transitions).

10 Draw labelled optical diagrams of single-beam and double-beam UV-Visible spectrophotometers and explain sources, monochromators, cells and detectors.

11 Perform quantitative analysis: single-component analysis (calibration-curve method), A(1%,1cm) method, and multi-component analysis by the simultaneous-equations method with derivation.

4.1 What is Spectroscopy? (Absolute Zero Start)

Spectroscopy is the science of studying the **interaction between matter and electromagnetic radiation**.

Definition: Spectroscopy is the branch of science that deals with the study of absorption, emission or scattering of electromagnetic radiation by matter, and uses the resulting spectrum to identify or quantify substances.

Simple Analogy

Think of **white light passing through a prism** to give seven colours (VIBGYOR). This is a spectrum. In a pharmacy laboratory, instruments do the same job more precisely: they shine light on a medicine sample and study what happens. From this study, we can answer two questions:

1 **What is this substance?** (qualitative analysis — identification)

2 **How much of it is present?** (quantitative analysis — assay)

Pharmacy relevance: A tablet labelled "Paracetamol 500 mg" — does it really contain 500 mg? Spectroscopy helps the quality-control analyst verify the exact amount.

4.2 Electromagnetic Radiation — The Wave and the Particle

Definition — Electromagnetic radiation (EMR): Energy that travels through space in the form of waves, consisting of an electric field and a magnetic field vibrating at right angles to each other and to the direction of travel. Light, radio waves, X-rays are all forms of EMR. It needs no medium to travel.

Wave Properties of Radiation

Definition — Wavelength (λ): The distance between two consecutive crests (peaks) of a wave. It is measured in nanometres (nm) in spectroscopy. Remember: 1 nm = 10^{-9} m.

Definition — Frequency (ν): The number of waves passing a fixed point in one second. Unit: hertz (Hz) or s^{-1}. More waves per second = higher frequency.

Definition — Wavenumber ($\bar{\nu}$): The number of waves per centimetre. Unit: cm^{-1}. Wavenumber is the reciprocal of wavelength:

Wavenumber ($\bar{\nu}$) = 1 / λ

Wavenumber is the favourite unit of IR spectroscopists because it is directly proportional to energy.

Definition — Amplitude (A): The height of the wave from its centre line to the peak. Amplitude is related to the *intensity* (brightness) of radiation, not its energy.

Definition — Energy (E): Each unit of radiation carries energy. The relationship is:

E = hν = hc / λ

where h = Planck's constant (6.626×10^{-34} J·s) and c = speed of light (3×10^{8} m/s).

Key relation: Energy is **directly proportional to frequency** and **inversely proportional to wavelength**. Short wavelength → high energy. Long wavelength → low energy.

Analogy — Sea Waves

- **Wavelength:** distance between two wave crests at the beach.
- **Frequency:** how many waves hit the shore per minute.
- **Amplitude:** how tall each wave is (small ripple vs huge wave).
- Fast, short waves crash harder (more energy) than slow, long, lazy waves.

Particle Nature of Radiation — The Photon

Definition — Photon: The smallest, indivisible packet (quantum) of electromagnetic radiation energy. Radiation is also a stream of tiny energy particles called photons.

Dual nature: Radiation behaves as a **wave** when travelling through space, but exchanges energy with matter as **particles** (photons). When a molecule absorbs light, it swallows whole photons — one photon at a time.

Mnemonic for wave–particle: "Wave on the Way, Particle at the Party" — radiation travels as a wave on the way, and behaves as a particle when it meets matter (at the "party").

Infographic Concept Box — Radiation Cheat Card

Property	Symbol	Unit	Relation to energy
Wavelength	λ	nm (metres)	Inverse ($E \propto 1/\lambda$)
Frequency	ν	Hz (s^{-1})	Direct ($E \propto \nu$)
Wavenumber	$\bar{\nu}$	cm^{-1}	Direct ($E \propto \bar{\nu}$)
Amplitude	A	—	Relates to intensity, not energy
Energy	E	Joules	$E = h\nu = hc/\lambda$

One-minute takeaway: $E = h\nu = hc/\lambda$. Short waves = high energy; long waves = low energy.

4.3 The Electromagnetic Spectrum

Definition — Electromagnetic spectrum: The arrangement of all types of electromagnetic radiation in order of increasing wavelength (or decreasing frequency/energy).

Region	Wavelength range	Energy	Spectroscopic technique	What it causes
Gamma rays	0.0001–0.01 nm	Very high	Mössbauer / γ spectroscopy	Nuclear transitions
X-rays	0.01–10 nm	High	X-ray spectroscopy	Inner-electron transitions
Vacuum UV	10–180 nm	High	— (absorbed by air)	Electronic transitions
Ultraviolet (UV)	**180–400 nm**	Moderate	**UV spectroscopy**	Electronic transitions
Visible	**400–800 nm**	Moderate	**Visible spectroscopy**	Electronic transitions
Infrared (IR)	**0.8 µm–200 µm** (800 nm–200,000 nm)	Low	**IR spectroscopy**	Molecular vibrations

Region	Wavelength range	Energy	Spectroscopic technique	What it causes
Microwave	0.1 mm–1 cm	Very low	Microwave spectroscopy	Molecular rotations
Radio waves	1 cm–meters	Lowest	NMR spectroscopy	Nuclear spin transitions

Mnemonic for order (shortest → longest wavelength): "Good X-ray Doctors Never Use UV Visible Instruments In Messy Rooms" — **G**amma, **X**-ray, **D**eep UV, **N**ear UV, **V**isible, **I**nfrared, **M**icrowave, **R**adio.

Order of increasing wavelength: Gamma < X-ray < UV < Visible < IR < Microwave < Radio **Order of increasing energy (reverse):** Radio < Microwave < IR < Visible < UV < X-ray < Gamma

Flowchart — Spectrum → Technique

```
EMR region (wavelength)  →  What it disturbs in the molecule  →  Technique
Gamma (10⁻⁵ nm)          →  Nucleus                           →  γ-spectroscopy
X-ray (10⁻² nm)          →  Inner electrons                   →  X-ray
UV-Visible (200–800 nm)  →  Outer (bonding) electrons         →  UV-Visible ✓
IR (2.5–25 µm)           →  Bonds vibrate like springs        →  IR ✓
Radio (cm–m)             →  Nuclei flip in magnetic field     →  NMR
```

Key takeaway: In this unit we study UV-Visible (electronic transitions) and IR (molecular vibrations) only.

4.4 Interaction of Radiation with Matter

When radiation meets matter, three things can happen:

1. Absorption Definition: The molecule takes in a photon and jumps from a lower energy state to a higher energy state. A photon is absorbed only if its energy exactly matches the energy gap between the two states. The rest of the light passes through — the absorbed colour is "missing" from the transmitted beam.

2. Emission Definition: An excited molecule falls back to a lower energy state, throwing out a photon. (Example: fluorescence, flame photometry.)

3. Scattering Definition: The photon hits the molecule and bounces off in a new direction, with or without a small change in its energy. (Example: Raman scattering, turbidity/nephelometry.)

One-line memory: "Matter either swallows (absorbs), spits (emits) or bounces (scatters) light."

4.5 UV-Visible Spectroscopy — Introduction

Definition: UV-Visible spectroscopy is the analytical technique that measures the absorption of ultraviolet (180–400 nm) and visible (400–800 nm) radiation by a sample, used for identification and quantitative analysis.

What happens inside the molecule: Electrons in the outer shells of atoms (bonding and non-bonding electrons) absorb UV-Visible photons and get promoted to higher energy orbitals. These are called

electronic transitions.

Why is it useful? Most drug molecules have structures (double bonds, benzene rings, carbonyl groups) that absorb UV light strongly. Each drug has a characteristic absorption pattern (spectrum), and the amount of absorption tells us the concentration.

India context: In Indian pharmaceutical QC laboratories, UV-Visible spectrophotometers are the most common instruments — the Indian Pharmacopoeia lists hundreds of UV assays (e.g., paracetamol tablets assayed at 257 nm). Every formulation QC lab in India, from small units in Baddi to large plants in Hyderabad, runs UV assays daily.

4.6 Beer–Lambert Law — Full Derivation (Step by Step)

This is the **single most important law** in UV-Visible spectroscopy. It combines two laws.

4.6.1 Lambert's Law

Definition (Lambert's law): When monochromatic (single-wavelength) light passes through a transparent absorbing medium, the decrease in intensity of light is proportional to the thickness of the medium, and each layer of equal thickness absorbs an equal fraction of the light entering it.

Starting point — the basic differential relation: Consider a thin layer of solution of thickness **dl**. Light of intensity **I** enters this layer. The tiny decrease in intensity **dI** across this layer is proportional to the incoming intensity **I** and the layer thickness **dl**:

dI = −k·I·dl ...(1)

The minus sign is used because intensity **decreases** as the light travels.

Step 1 — Rearrange: Divide both sides by I: **dI / I = −k·dl** ...(2)

Step 2 — Integrate: Integrate from the starting intensity $\mathbf{I_0}$ (before entering the solution) to the final intensity **I** (after travelling path length **b**):

$\int(I_0 \to I)\, dI/I = -k \int(0 \to b)\, dl$

Step 3 — Solve the integrals: Left side: $\ln I - \ln I_0 = \ln(I/I_0)$ Right side: $-k \cdot b$

So: $\mathbf{\ln(I/I_0) = -k \cdot b}$, or $\mathbf{I = I_0 \cdot e^{(-k \cdot b)}}$

This is the **exponential form** — Lambert's law.

In words: The intensity of light falls exponentially with the thickness of the absorbing medium.

4.6.2 Beer's Law

Definition (Beer's law): When monochromatic light passes through a solution, the decrease in intensity of light is proportional to the concentration of the absorbing substance, and each equal concentration absorbs an equal fraction of light.

Starting again from the differential relation, now the constant k is proportional to the concentration **c**:

dI = −k'·c·I·dl ...(3)

Following the same steps (divide by I, integrate), we get:

$\ln(I/I_0) = -k' \cdot c \cdot b$, or **$I = I_0 \cdot e^{(-k' \cdot c \cdot b)}$**

4.6.3 Combining Both — Beer–Lambert Law

Both laws say the same thing in different words: intensity falls exponentially with path length **b** and concentration **c**. Combining:

$\ln(I/I_0) = -\varepsilon \cdot b \cdot c$ (natural-log form)

Now convert the natural logarithm to base-10 logarithm. Since $\ln(x) = 2.303 \cdot \log(x)$:

$2.303 \cdot \log(I/I_0) = -\varepsilon \cdot b \cdot c$

Divide by -2.303 and define the **molar absorptivity (ε)**:

$A = -\log(I/I_0) = \log(I_0/I) = \varepsilon \cdot b \cdot c$

The Beer–Lambert Law (final form):

$A = \varepsilon \cdot b \cdot c$

4.6.4 Terms of the Equation (Know Every Term)

Term	Name	Meaning	Units
A	Absorbance (optical density)	$\log(I_0/I)$; measures how much light is absorbed	Unitless
ε	Molar absorptivity (molar extinction coefficient)	Absorbance of 1 mol/L solution in a 1 cm cell; a characteristic constant of the substance at a given wavelength	$L \cdot mol^{-1} \cdot cm^{-1}$
b	Path length	Thickness of solution the light passes through (usually the cell)	cm (usually 1 cm)
c	Concentration	Concentration of absorbing substance	mol/L (for ε)

Definition — Absorbance (A): $A = \log(I_0/I) = -\log(I/I_0)$. It is unitless. A = 0 means no absorption; A = 1 means 90% of light absorbed.

Definition — Molar absorptivity (ε): The absorbance of a 1 molar (1 mol/L) solution measured in a 1 cm path-length cell at a particular wavelength. It tells how strongly the substance absorbs — like the "absorption fingerprint strength" of the drug.

Flowchart — Derivation in 5 steps:

```
1. START:  dI = −k·I·dl   (intensity falls through a thin layer)
2. DIVIDE: dI/I = −k·dl
3. INTEGRATE: ln(I/I₀) = −k·b     →  I = I₀·e^(−k·b)
4. CONVERT: ln → log₁₀ using ln(x) = 2.303·log(x)
5. COMBINE Beer + Lambert:  A = ε·b·c   ✓
```

Key takeaway: Absorbance is directly proportional to concentration and path length. Double the concentration → double the absorbance.

Common Mistake

Wrong: "Absorbance has units." **Correct:** Absorbance is unitless (a logarithm). Only ε carries units ($L \cdot mol^{-1} \cdot cm^{-1}$).

4.7 Transmittance vs Absorbance

Definition — Transmittance (T): The fraction of incident light that passes through the sample:

$T = I / I_0$

Definition — Percent transmittance (%T): $\%T = T \times 100 = (I/I_0) \times 100$.

Relation between A and T — Derivation

Start from absorbance: $A = -\log_{10}(T)$ ...(1)

Convert to %T. Since $T = \%T/100$:

$A = -\log_{10}(\%T/100)$ $A = -[\log_{10}(\%T) - \log_{10}(100)]$ $A = -\log_{10}(\%T) + \log_{10}(100)$ $A = -\log_{10}(\%T) + 2$

$A = 2 - \log(\%T)$ ✓

Memorise: T = 1 (100%T) → A = 0; T = 0.1 (10%T) → A = 1; T = 0.01 (1%T) → A = 2.

Absorbance (A)	Transmittance (T)	%T
0	1.00	100%
0.5	0.316	31.6%
1.0	0.10	10%
2.0	0.01	1%

CALCULATION BOX 1 — Beer–Lambert concentration calculation

Question: A paracetamol solution shows absorbance A = 0.600 at 257 nm in a 1 cm cell. The molar absorptivity ε of paracetamol at 257 nm is 6,700 $L \cdot mol^{-1} \cdot cm^{-1}$. Calculate the molar concentration.

Solution: Step 1: Write the law: $A = \varepsilon \cdot b \cdot c \rightarrow c = A / (\varepsilon \cdot b)$ Step 2: Substitute: $c = 0.600 / (6{,}700 \times 1)$ Step 3: $c = 0.600 / 6{,}700 = 8.96 \times 10^{-5}$ mol/L **Answer:** $c = 8.96 \times 10^{-5}$ mol/L

CALCULATION BOX 2 — %T ↔ A conversions

(a) A sample transmits 25% of incident light. Find A. Step 1: $A = 2 - \log(\%T)$ Step 2: $A = 2 - \log(25) = 2 - 1.398 = 0.602$ **Answer:** A = 0.602

(b) A sample has A = 1.30. Find %T. Step 1: $1.30 = 2 - \log(\%T) \rightarrow \log(\%T) = 2 - 1.30 = 0.70$ Step 2: %T = 10^0.70 = 5.01 **Answer:** %T = 5.01% (≈5%)

4.8 Deviations from Beer's Law

The law is followed only under certain conditions. Deviations fall into three types.

Type 1 — Chemical Deviations

Definition: Occur when the absorbing substance changes its chemical form (dissociation, association, reaction with solvent) with concentration.

Examples:

- A weak acid drug absorbs differently in dissociated vs undissociated form; changing concentration or pH shifts the equilibrium → non-linearity.
- Association of molecules at high concentration (dimer formation).

Control: Use buffered solutions at constant pH.

Type 2 — Instrumental Deviations

Definition: Occur because of instrument limitations — mainly when the light is not truly monochromatic (stray light, wide slit width).

Examples:

- Stray light (light of wrong wavelength reaching the detector) causes positive deviation, especially at high absorbance.
- Wide slit width → polychromatic light → curved calibration plot.

Control: Use narrow slit widths, keep absorbance below ~1.5, calibrate instruments regularly.

Type 3 — Fundamental Deviations

Definition: Occur at very high concentrations (> 0.01 mol/L) because the law is a limiting law valid only for dilute solutions; also when the refractive index of the solution changes.

Example: A drug solution at 0.1 M shows a curved calibration line because molecules interact with each other and the average environment of each molecule changes.

Control: Dilute the sample so concentration stays within the linear range.

Common Confusion — Deviations vs Errors: Deviations are systematic departures from the linear law (a property of the system), while errors are mistakes made during measurement (e.g., dirty cells, wrong wavelength). Diluting a too-concentrated sample fixes fundamental deviation; cleaning cells fixes error.

One-minute takeaway: "Chemistry (species changes), Instrument (stray light), Fundamental (too concentrated)" — the three families of deviation.

4.9 Electronic Transitions

Definition: When a molecule absorbs UV-Visible light, an electron jumps from a lower-energy molecular orbital to a higher-energy (antibonding) orbital. This jump is an electronic transition.

Orbitals involved: σ (bonding), π (bonding), n (non-bonding lone pair), π *(antibonding), σ* (antibonding).

The Four Transitions (in order of increasing energy required)

1. **n → π*** — non-bonding electron → π* orbital. **Lowest energy.** Wavelength ~270–350 nm (UV). Example: carbonyl C=O groups (ketones like camphor).
2. **π → π*** — bonding π electron → π* orbital. Moderate energy. Wavelength ~180–260 nm (UV). Example: alkenes, benzene ring.
3. **n → σ*** — non-bonding electron → σ* orbital. High energy. Wavelength ~150–200 nm (vacuum UV).
4. **σ → σ*** — bonding σ electron → σ* orbital. **Highest energy.** Wavelength <150 nm (vacuum UV). Example: saturated alkanes.

Order of energy: n→π < *π→π* < n→σ < *σ→σ*

Order of wavelength (reverse): n→π *(longest)* > *π→π* > n→σ > *σ→σ* (shortest)

Which are useful in UV-Visible (200–800 nm)? nπ and ππ* — these need the least energy and appear in the normal UV-visible range. σσ and nσ* need very high energy, fall in vacuum UV (absorbed by air and quartz), and are rarely useful for pharmacy.

Analogy: Like jumping between floors of a building — the basement-to-ground floor jump (n→π*) needs the least effort; the basement-to-roof jump (σ→σ*) needs the most.

4.10 Chromophores and Auxochromes

Definition — Chromophore: A group of atoms in a molecule responsible for absorbing UV-Visible radiation — the "colour-bearing" part of the molecule. A chromophore always has either multiple bonds (C=C, C=O) or atoms with lone pairs.

Examples of chromophores:

Chromophore	Group	Typical absorption
Ethylene	C=C	~175 nm (π→π*)
Carbonyl	C=O	~280 nm (n→π*)
Benzene ring	C_6H_6	~255 nm (π→π*, B-band)
Nitro	$-NO_2$	~270 nm (n→π*)
Azo	−N=N−	~350 nm (coloured!)
Conjugated diene	C=C−C=C	~217 nm (π→π*)

Definition — Auxochrome: A group that does **not** itself absorb UV-Visible light, but when attached to a chromophore, it changes (usually intensifies or shifts) the absorption. It is the "colour helper" — typically a group with lone pairs (−OH, $-NH_2$, $-OCH_3$, −SH) or charged groups.

Examples of auxochromes: −OH (hydroxyl), $-NH_2$ (amino), $-OCH_3$ (methoxy), −SH (thiol), −Cl, −COOH.

Effect of auxochromes: Usually cause a **bathochromic shift** (toward longer wavelength) and a **hyperchromic effect** (increase in intensity).

Absorption Bands of Benzene (K, R, B, E bands)

Band	Wavelength	Transition	Intensity	Feature
K-band (konjugierte)	~217 nm	π→π* (conjugated)	High (strong)	Conjugated systems; moves with substitution
E-band	~180–200 nm	π→π* (ethylenic)	Very high	Benzene ethylenic absorption
B-band (benzenoid)	~255 nm	π→π* (benzene)	Low (weak)	Fine structure of benzene ring
R-band (radikalartig)	~270–350 nm	n→π*	Very low	Carbonyl/nitro groups; solvent-sensitive

Mnemonic: "Kids Eat Big Red apples" — **K**, **E**, **B**, **R** bands.

Pharmacy relevance: Paracetamol has a benzene ring (B-band region) plus −OH and $-NHCOCH_3$ auxochromes; its strong absorption at 257 nm is what the Indian Pharmacopoeia assay uses.

4.11 Spectral Shifts

Definitions:

1. **Bathochromic shift (red shift):** Shift of λmax toward **longer** wavelength (lower energy). Remember: "batho" — think of going *deeper* (longer) into the spectrum.
2. **Hypsochromic shift (blue shift):** Shift of λmax toward **shorter** wavelength (higher energy).
3. **Hyperchromic effect: Increase** in absorption intensity (higher ε or A). "Hyper" = more.
4. **Hypochromic effect: Decrease** in absorption intensity (lower ε or A). "Hypo" = less.

Memory table:

Shift	What changes	Direction	Memory trick
Bathochromic	λmax	Longer λ	"Batho = Bath-tub long"
Hypsochromic	λmax	Shorter λ	"Hypso = Hip-shot short"
Hyperchromic	Intensity	Increases	"Hyper = High"
Hypochromic	Intensity	Decreases	"Hypo = Low"

4.12 Effect of Solvent on Absorption Spectra

The solvent can change both the position (λmax) and intensity of absorption bands.

n → π* transition (e.g., carbonyl C=O)

- In a **polar solvent** (e.g., water, ethanol): the solvent stabilises the ground state (non-bonding electrons) more than the excited state → energy gap **increases** → **hypsochromic shift** (shorter λ) and **hypochromic effect** (lower intensity).
- In a **non-polar solvent** (e.g., hexane): less stabilisation → absorption at longer λ.

π → π* transition (e.g., benzene, conjugated dienes)

- In a **polar solvent**: the excited state (more polar) is stabilised more than the ground state → energy gap **decreases** → **bathochromic shift** (longer λ) and **hyperchromic effect** (higher intensity).
- In a **non-polar solvent**: shorter λ.

Summary table:

Transition	Polar solvent effect	Non-polar solvent effect
n → π*	Hypsochromic (blue) shift, lower intensity	Longer λ, higher intensity
π → π*	Bathochromic (red) shift, higher intensity	Shorter λ, lower intensity

Mnemonic: "Polar pushes π→π *to the red, and n→π* to the blue."

Common confusion: Students mix up which transition shifts which way. Rule of thumb: the transition whose **excited state is more polar** (π→π*) gets red-shifted by polar solvents; the one whose **ground state has lone pairs that H-bond with solvent** (*n*→π) gets blue-shifted.

India/pharmacy relevance: Drug assays in the Indian Pharmacopoeia specify the exact solvent (e.g., 0.1 M NaOH, methanol) because λmax changes with solvent. Never substitute solvents in a pharmacopoeial assay.

4.13 Instrumentation of UV-Visible Spectrophotometer

A spectrophotometer has five main parts: **source → wavelength selector → sample holder → detector → readout**.

Flowchart — Light Path

```
Radiation source → Filter/Monochromator (selects λ) → Sample cell (absorbs some light)
→ Detector (converts light to electrical signal) → Amplifier → Meter/Display (reads A or %T)
```

4.13.1 Sources of Radiation

1. Hydrogen / Deuterium discharge lamp (UV region, 180–400 nm)

- A tube containing hydrogen or deuterium gas at low pressure; an electric discharge excites the gas, which emits UV radiation.
- Deuterium lamp gives higher intensity and longer life than hydrogen lamp — the standard UV source in modern instruments.

2. Tungsten filament lamp (Visible region, 350–800 nm)

- A tungsten wire heated electrically (like an ordinary bulb); emits continuous radiation in the visible range.
- Stable, inexpensive; the standard visible source.

3. Xenon arc lamp

- An intense electric arc between electrodes in xenon gas; covers both UV and visible ranges (200–1,000 nm).
- Very bright; used when high intensity is needed (e.g., fluorescence work).

Requirement of a good source: stable intensity, long life, and continuous radiation over the required wavelength range.

4.13.2 Wavelength Selectors

A. Filters

- Simple glass pieces that absorb all wavelengths except one narrow band.
- Cheap but not precise; used in simple colorimeters, not in spectrophotometers.

B. Monochromators Definition: A device that isolates a single (narrow band of) wavelength from the broad radiation of the source.

Prism monochromator:

- A glass or quartz prism bends (refracts) different wavelengths by different amounts, spreading light into a spectrum; a slit selects one wavelength.
- Simple and cheap, but dispersion is non-linear (wavelengths crowd together at one end).

Diffraction grating monochromator:

- A surface with thousands of closely spaced lines (grooves) per cm; light diffracts (bends around the lines) and each wavelength emerges at a different angle; a slit selects the desired wavelength.
- Dispersion is linear and resolution is much better than a prism — the standard in modern instruments.

C. Slits

- Narrow openings (entrance slit and exit slit) that control the band of wavelengths passing through and the intensity reaching the detector.
- Narrower slit = purer wavelength but weaker signal.

4.13.3 Sample Cells (Cuvettes)

Definition: Transparent containers holding the sample solution in the light path, usually 1 cm path length.

- **Quartz cells:** Used for **UV region** — quartz transmits UV light (glass does not).
- **Glass cells:** Used for **visible region** only — glass absorbs UV below ~350 nm.

Handling rules: cells must be clean (no fingerprints), filled to the correct level, and the reference cell (with pure solvent = blank) must be matched to the sample cell.

Common mistake: Using glass cells for UV measurements below 350 nm — glass absorbs there and ruins the reading. Always use **quartz for UV**.

4.13.4 Detectors

1. Phototube (photoemissive cell)

- **Construction:** An evacuated glass tube containing a photosensitive cathode (coated with alkali metal) and an anode.
- **Working:** Light strikes the cathode → electrons are ejected (photoelectric effect) → collected by the anode → small electric current proportional to light intensity.

2. Photomultiplier tube (PMT)

- **Construction:** Like a phototube, but with a series of 9–13 extra electrodes called **dynodes** arranged in a cascade.
- **Working:** Each electron ejected from the cathode strikes the first dynode, knocking out 2–3 secondary electrons; these strike the next dynode, multiplying at each stage (total multiplication ~10^6). Result: an enormous amplified current.
- **Advantage:** Extremely sensitive — detects very faint light; the standard detector in UV-Visible instruments.

3. Photovoltaic cell (barrier-layer cell)

- **Construction:** A semiconductor layer (e.g., selenium) coated on an iron plate, covered with a thin transparent metal film.
- **Working:** Light generates an electric voltage directly (no external battery needed) — the current flows and is measured.
- **Advantage:** Simple, rugged, cheap; needs no power supply. Less sensitive than PMT.

4. Silicon photodiode

- **Construction:** A silicon p–n junction semiconductor.
- **Working:** Light falling on the junction creates electron–hole pairs → current proportional to light intensity (with a small reverse bias applied).
- **Advantage:** Small, fast, sensitive over a wide range; widely used in modern diode-array instruments.

4.13.5 Types of Instruments — Single-Beam and Double-Beam

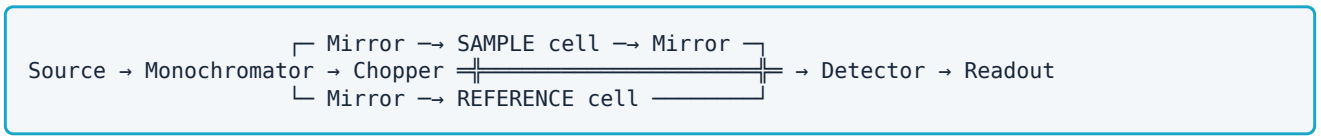
Single-beam spectrophotometer:

- **Working:** Light passes through ONE path. First the blank (solvent) is placed and the instrument is set to zero absorbance; then the blank is removed and the sample is placed and its absorbance read.
- **Disadvantage:** Two separate measurements; if the source intensity drifts between blank and sample readings, error occurs.

Double-beam spectrophotometer:

- **Working:** A rotating mirror/chopper splits the beam into TWO paths — one through the sample cell, one through the reference (blank) cell — and the detector measures both alternately, continuously correcting for source fluctuations.
- **Advantage:** Automatic blank correction, faster, more accurate; used for scanning full spectra.

Double-beam optical diagram (textual):



(In a real diagram: Source → Entrance slit → Dispersing element → Exit slit → Beam splitter/chopper → Sample and Reference beams → Detector → Amplifier → Display.)

India context: Most Indian college and QC laboratories use double-beam instruments (e.g., Shimadzu, Systronics brands commonly sold in India) because they allow direct spectral scanning and are now affordable.

4.14 Quantitative Analysis — Single-Component Analysis

When only ONE absorbing drug is present, we can find its concentration in two ways:

Method 1 — Calibration-Curve Method (most common)

Steps:

1. Prepare standard solutions of the pure drug of known concentrations (e.g., 2, 4, 6, 8, 10 µg/mL).
2. Measure absorbance of each at λmax; also measure a blank (solvent) and set zero.
3. Plot a graph: **absorbance (Y-axis) vs concentration (X-axis)** → a straight line through the origin (Beer's law).
4. Measure the absorbance of the unknown sample; read its concentration from the graph (interpolate).

CALCULATION BOX 3 — Calibration-curve interpolation

Question: Standard paracetamol solutions gave: 2 µg/mL → A 0.20; 4 µg/mL → A 0.40; 6 µg/mL → A 0.61; 8 µg/mL → A 0.80; 10 µg/mL → A 1.00. An unknown tablet solution shows A = 0.52. Find its concentration.

Solution: Step 1: The plot of A vs c is a straight line with slope ≈ 0.10 per µg/mL (since 10 µg/mL gives A 1.00). Step 2: For the unknown: c = A / slope = 0.52 / 0.10 = 5.2 µg/mL. (From the graph, A 0.52 falls between the 4 and 6 µg/mL points, at 5.2 µg/mL.) **Answer:** 5.2 µg/mL.

Method 2 — A(1%, 1cm) Method (specific absorbance)

Definition — A(1%,1cm): The absorbance of a 1% w/v solution (1 g per 100 mL) measured in a 1 cm cell at a specified wavelength. It is a pharmacopoeial constant for each drug.

Formula: c (in g/100 mL) = A / A(1%,1cm)

CALCULATION BOX 4 — A(1%,1cm) calculation

Question: The A(1%,1cm) of aspirin at 278 nm is 17. An aspirin tablet solution (after dilution) shows A = 0.340. Calculate the concentration in g/100 mL and in mg/mL.

Solution: Step 1: c (g/100 mL) = A / A(1%,1cm) = 0.340 / 17 = 0.020 g/100 mL. Step 2: Convert: 0.020 g/100 mL = 20 mg/100 mL = 0.20 mg/mL. **Answer:** 0.020 g/100 mL = 0.20 mg/mL.

Pharmacy relevance: The Indian Pharmacopoeia gives A(1%,1cm) values for many drugs — the QC analyst needs no standard of the pure drug, just this constant.

4.15 Multi-Component Analysis — Simultaneous-Equations Method

When TWO absorbing drugs (x and y) are mixed together, each absorbs at both wavelengths. We measure at two wavelengths and solve two equations.

Derivation (step by step)

Step 1 — Write Beer's law for component x alone: At wavelength λ1: A1x = a1x·b·Cx ...(i) At wavelength λ2: A2x = a2x·b·Cx ...(ii) where a1x = absorptivity of x at λ1, a2x = absorptivity of x at λ2, Cx = concentration of x.

Step 2 — Write Beer's law for component y alone: At λ1: A1y = a1y·b·Cy ...(iii) At λ2: A2y = a2y·b·Cy ...(iv)

Step 3 — Absorbances are additive (mixture): Total absorbance at λ1 = A1 = A1x + A1y Total absorbance at λ2 = A2 = A2x + A2y

Step 4 — Substitute (i)–(iv): A1 = a1x·b·Cx + a1y·b·Cy ...(v) **A2 = a2x·b·Cx + a2y·b·Cy** ...(vi)

Step 5 — Take b = 1 cm (standard cells): A1 = a1x·Cx + a1y·Cy ...(vii) A2 = a2x·Cx + a2y·Cy ...(viii)

Step 6 — Solve the two equations simultaneously for Cx and Cy (by elimination or substitution; worked below in the Calculation Box).

Key assumption: The two components do not chemically interact, and Beer's law holds for each at both wavelengths.

When to choose λ1 and λ2: λ1 = λmax of drug x, λ2 = λmax of drug y.

CALCULATION BOX 5 — Two-component simultaneous equations (full solution)

Question: A mixture contains drugs X and Y. Absorptivities (1% , 1 cm): at λ1 (λmax of X): a1x = 120, a1y = 40; at λ2 (λmax of Y): a2x = 50, a2y = 200. The mixture (1 cm cell) shows A1 = 0.80 and A2 = 0.90. Calculate Cx and Cy (in g/100 mL).

Solution: Step 1: Write the two equations (b = 1 cm): 0.80 = 120·Cx + 40·Cy ...(vii) 0.90 = 50·Cx + 200·Cy ...(viii)

Step 2: Eliminate Cy. Multiply (vii) by 5: 4.00 = 600·Cx + 200·Cy Step 3: Subtract (viii) from it: 4.00 − 0.90 = (600 − 50)·Cx 3.10 = 550·Cx → Cx = 3.10 / 550 = 0.005636 g/100 mL

Step 4: Substitute Cx in (vii): 0.80 = 120(0.005636) + 40·Cy 0.80 = 0.6764 + 40·Cy → 40·Cy = 0.1236 → Cy = 0.00309 g/100 mL

Answer: Cx = 0.00564 g/100 mL; Cy = 0.00309 g/100 mL.

Pharmacy relevance: A combination tablet (e.g., paracetamol + ibuprofen, or aspirin + paracetamol) can be assayed without separating the two drugs — the classic QC application of this method.

Common Mistake

Wrong: Measuring both wavelengths at the same λmax. **Correct:** λ1 and λ2 should be the λmax of each component — the equations fail if the two wavelengths are too close (absorptivities become nearly equal and the solution is unstable).

4.16 Applications of UV-Visible Spectroscopy in Pharmacy

1. **Quantitative analysis (assay):** Determining drug content in tablets, capsules, injections (e.g., paracetamol tablets at 257 nm, aspirin at 278 nm).
2. **Identification:** Comparing the UV spectrum (λmax and shape) of a sample with that of a reference standard.
3. **Multi-component analysis:** Simultaneous estimation of two drugs in combination products (simultaneous-equations method).
4. **Dissolution testing:** Monitoring drug release from tablets over time.
5. **Purity checking:** Extra absorption bands reveal impurities.
6. **Determination of dissociation constants (pKa)** of drugs by measuring spectra at different pH values.
7. **Reaction kinetics:** Following a reaction by the change in absorbance with time.

Infographic Concept Box — Chapter 4 One-Page Map

```
ELECTROMAGNETIC RADIATION
├─ Wave: λ, ν, ν̄, amplitude | Particle: photon | E = hν = hc/λ
├─ Spectrum order: Gamma < X-ray < UV < Visible < IR < Microwave < Radio
├─ Interaction: ABSORB (in) · EMIT (out) · SCATTER (bounce)
└─ UV-VISIBLE (200–800 nm): ELECTRONIC transitions
    ├─ Law: A = εbc  (from dI = −kIdl → integrate → convert ln→log)
    ├─ A = 2 − log(%T)
    ├─ Deviations: Chemical · Instrumental · Fundamental
    ├─ Transitions (energy): n→π* < π→π* < n→σ* < σ→σ*
    ├─ Chromophore (absorbs) + Auxochrome (shifts/intensifies)
    ├─ Bands: K · E · B · R  |  Shifts: batho/hypso (λ) · hyper/hypo (intensity)
    ├─ Solvent: polar → π→π* red shift · n→π* blue shift
    ├─ Instrument: Source → Monochromator → Cell → Detector → Readout
    └─ Analysis: calibration curve · A(1%,1cm) · simultaneous equations
```

Key Points — Chapter 4

1. Spectroscopy studies the interaction of matter with electromagnetic radiation.
2. E = hν = hc/λ: short wavelength = high energy.
3. Wavelength order: gamma < X-ray < UV < visible < IR < microwave < radio.
4. Beer–Lambert law: A = εbc; derived from dI = −k·I·dl by integration and log conversion.
5. A = 2 − log(%T).
6. Deviations: chemical (species change), instrumental (stray light), fundamental (too concentrated).
7. Transition energy order: n→π < *π→π* < n→σ < *σ→σ*; only n→π *and* *π→π* are analytically useful.
8. Chromophore absorbs; auxochrome modifies. K/E/B/R bands describe benzene absorptions.

9 Bathochromic = longer λ; hypsochromic = shorter λ; hyperchromic = more intense; hypochromic = less intense.

10 Polar solvents shift π→π *to red and n→π* to blue.

11 Deuterium lamp (UV), tungsten lamp (visible), grating monochromator, quartz cells for UV, PMT detector.

12 Single-beam: one path, blank then sample; double-beam: two paths, automatic correction.

13 Single-component: calibration curve or A(1%,1cm); two-component: simultaneous equations.

Common Confusions — Chapter 4

1 **Transmittance vs absorbance:** T = I/I_0 (fraction passing through); A = $\log(I_0/I)$ (fraction absorbed, log scale). They are inversely related, not simply opposite.

2 **Bathochromic vs hyperchromic:** bathochromic changes wavelength (λmax moves); hyperchromic changes intensity (peak height grows). A shift can be both at once.

3 **Chromophore vs auxochrome:** the chromophore absorbs light by itself; the auxochrome cannot absorb alone — it only modifies the chromophore's absorption.

4 **K-band vs R-band:** K-band is intense π→π *of conjugated systems; R-band is weak n→π* of carbonyl/nitro groups.

Common Mistakes — Chapter 4

1 Giving absorbance units — absorbance is unitless.

2 Using glass cells below 350 nm — glass absorbs UV; use quartz.

3 Forgetting the blank — always zero the instrument with pure solvent first.

4 Measuring too-concentrated solutions (A > 2) — dilute into the linear range.

5 Choosing two wavelengths too close together in simultaneous equations.

Mnemonics — Chapter 4

- Spectrum order: **"Good X-ray Doctors Never Use UV Visible Instruments In Messy Rooms"**
- Wave–particle: **"Wave on the Way, Particle at the Party"**
- Bands: **"Kids Eat Big Red apples"** (K, E, B, R)
- Interaction: **"Swallow, Spit, Bounce"** (absorb, emit, scatter)
- Solvent: **"Polar pushes π→π *to the red, n→π* to the blue"**

Exam Summary — Chapter 4

- EMR has wave properties (λ, ν, $\bar{\nu}$, amplitude) and particle nature (photon); E = hν = hc/λ.

- Spectrum order by increasing wavelength: gamma, X-ray, UV, visible, IR, microwave, radio.
- Beer–Lambert law derivation: dI = −k·I·dl → integrate → I = I_0e^(−kbc) → A = εbc; A = 2 − log(%T).
- Deviations: chemical, instrumental, fundamental — with one example each.
- Electronic transitions in energy order: n→π < *π→π* < n→σ < *σ→σ*.
- Chromophores (C=C, C=O, benzene, −NO_2, −N=N−), auxochromes (−OH, −NH_2, −OCH_3), K/E/B/R bands.
- Shifts: bathochromic (longer λ), hypsochromic (shorter λ), hyperchromic (↑intensity), hypochromic (↓intensity); solvent effects table.
- Instrumentation: sources (deuterium, tungsten, xenon), monochromators (prism, grating), quartz vs glass cells, detectors (phototube, PMT, photovoltaic, photodiode), single-beam vs double-beam diagrams.
- Quantitation: calibration curve, A(1%,1cm), simultaneous-equations derivation for two components.

One-Minute Revision — Chapter 4

A = εbc. E = hc/λ. Order: gamma→radio (longer λ, lower E). Transitions: n→π* lowest energy. Batho = long λ, hypso = short λ, hyper = more intense, hypo = less intense. Quartz for UV, PMT detector, grating monochromator. Blank first, then sample. Dilute if A > 1.5.

Practice Numericals — Chapter 4 (unworked, answers given)

1 A solution of drug X (ε = 12,500 L·mol^{-1}·cm^{-1}) in a 1 cm cell shows A = 0.75. Find its molar concentration. **Answer: 6.0 × 10^{-5} mol/L**

2 Convert A = 0.43 to %T. **Answer: 37.2%**

3 A mixture gives A1 = 0.65, A2 = 0.55 with absorptivities a1x = 100, a1y = 30, a2x = 45, a2y = 150 (1 cm cell). Find Cx, Cy in g/100 mL. **Answer: Cx = 0.00591 g/100 mL, Cy = 0.00190 g/100 mL**

Exam Questions — Chapter 4

Short answers (2–5 marks):

1 Define spectroscopy, photon and wavenumber.

2 State Beer's law and Lambert's law.

3 Derive the relation A = 2 − log(%T).

4 Arrange n→π, *π→π*, n→σ, *σ→σ* in order of energy and state which are useful in UV-Visible spectroscopy.

5 Define chromophore and auxochrome with two examples each.

6 What is a bathochromic shift? How does a polar solvent affect n→π* transitions?

7 Why are quartz cells used for UV measurements?

8 What is A(1%,1cm)? Write its formula.

Long answers (10–15 marks):

1 Derive the Beer–Lambert law starting from dI = −k·I·dl. Show all steps including integration and the conversion from natural to base-10 logarithm.

2 Discuss the three types of deviations from Beer's law with examples and the precautions to avoid each.

3 Describe the instrumentation of a UV-Visible spectrophotometer: sources, wavelength selectors, sample cells and detectors. Draw labelled optical diagrams of single-beam and double-beam instruments and compare them.

4 Derive the simultaneous-equations method for the analysis of a two-component mixture. Work out a numerical example.

5 Explain chromophores, auxochromes, the K/E/B/R bands and the four spectral shifts, with the effect of solvents on n→π *and π→π* transitions.

MCQs (answer key at end):

1 The Beer–Lambert law is obeyed best at: (a) very high concentration (b) dilute solutions (c) solid samples (d) any concentration → **(b)**

2 A = 1 corresponds to %T of: (a) 100 (b) 50 (c) 10 (d) 1 → **(c)**

3 Which transition requires the least energy? (a) σ→σ *(b) n→σ* (c) π→π *(d) n→π* → **(d)**

4 Bathochromic shift means: (a) shift to shorter wavelength (b) shift to longer wavelength (c) increase in intensity (d) decrease in intensity → **(b)**

5 $-NH_2$ attached to benzene is an example of: (a) chromophore (b) auxochrome (c) fluorophore (d) none → **(b)**

6 For UV measurements below 350 nm, cells must be made of: (a) glass (b) plastic (c) quartz (d) metal → **(c)**

7 The most sensitive UV detector is the: (a) photovoltaic cell (b) phototube (c) photomultiplier tube (d) thermocouple → **(c)**

8 A polar solvent shifts a π→π* band toward: (a) shorter λ (b) longer λ (c) no change (d) zero absorbance → **(b)**

Viva questions:

1 What does a double-beam instrument correct automatically? (Source fluctuations / drift.)

2 Why is absorbance unitless? (It is a logarithm of a ratio.)

3 Which lamp would you switch on for a measurement at 500 nm? (Tungsten lamp.)

4 Your calibration curve bends at high concentration — what do you do? (Dilute the sample.)

Application-based:

1 A QC analyst must assay paracetamol + ibuprofen combination tablets without separation. Which method and which measurements would you recommend? (Simultaneous-equations method at the λmax of each drug.)

2 The IP monograph specifies methanol as solvent for a UV assay. A trainee uses water instead and λmax shifts. Explain why. (Solvent polarity changes the energy gaps of n→π *and* π→π transitions.)

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CHAPTER 5 — INFRARED SPECTROSCOPY AND FTIR

Learning Objectives

After studying this chapter, the student will be able to:

1 Define IR spectroscopy and name the near, mid and far IR regions with their ranges.

2 Describe the fundamental modes of vibration in polyatomic molecules (stretching: symmetric/asymmetric; bending: scissoring, rocking, wagging, twisting) and apply the 3n−6 / 3n−5 rule.

3 State Hooke's-law equation for vibrational frequency and explain the meaning of each term.

4 Explain how reduced mass and force constant affect vibrational frequency (C−H vs C−D; single vs double vs triple bonds).

5 Draw the optical diagram of a dispersive IR spectrophotometer and explain the source, monochromator and detectors (thermocouple, bolometer, pyroelectric).

6 Compare sample-handling techniques: Nujol mull, KBr pellet, thin film and ATR.

7 Explain the FTIR principle: Michelson interferometer, interferogram, Fourier transform, and the Fellgett (multiplex) and Jacquinot (throughput) advantages.

8 List pharmaceutical applications: identification/fingerprint region, polymorphism detection, purity checking.

5.1 Introduction to IR Spectroscopy (Absolute Zero Start)

Definition: Infrared (IR) spectroscopy is the analytical technique that measures the absorption of infrared radiation (wavelength ~0.8 μm to 200 μm; wavenumber 12,800–50 cm^{-1}) by a sample, used mainly to

identify functional groups in molecules.

What happens inside the molecule?

In UV spectroscopy, electrons jump. In IR spectroscopy, **bonds vibrate**. Imagine each chemical bond as a tiny spring connecting two balls (atoms). The spring constantly stretches and bends. When IR light of exactly the right frequency hits the bond, the bond **absorbs** it and vibrates more strongly. Each type of bond (O−H, C=O, C−H) absorbs at a characteristic frequency — like each musical instrument having its own sound.

The Three IR Regions

Region	Wavenumber range	Wavelength range	Use
Near IR	12,800–4,000 cm^{-1}	0.8–2.5 μm	Overtones; quantitative analysis
Mid IR	4,000–400 cm^{-1}	2.5–25 μm	**Functional-group identification (most used)**
Far IR	400–50 cm^{-1}	25–200 μm	Heavy-atom vibrations, lattice modes

Memory: Mid IR (4,000–400 cm^{-1}) is the "workhorse" region — this is where pharmacists identify drugs.

Analogy: Pluck guitar strings of different thickness — thick strings (heavy atoms, weak bonds) give low notes; thin tight strings (light atoms, strong bonds) give high notes. IR "listens" to these molecular notes.

India context: Indian drug manufacturers use IR spectroscopy to confirm the identity of every raw material batch (e.g., incoming paracetamol powder) before manufacturing — the IR spectrum must match the reference spectrum in the Indian Pharmacopoeia.

5.2 Fundamental Modes of Vibration in Polyatomic Molecules

Definition — Molecular vibration: The periodic motion of atoms within a molecule — bonds stretching (changing length) and bending (changing angle).

Degrees of Freedom — How Many Vibrations?

Formula:

- **Non-linear molecule:** number of fundamental vibrations = **3n − 6**
- **Linear molecule:** number of fundamental vibrations = **3n − 5**

where n = number of atoms in the molecule.

Example: Water (H_2O, non-linear, n = 3): 3(3) − 6 = **3 vibrations**. CO_2 (linear, n = 3): 3(3) − 5 = **4 vibrations**.

Types of Vibrations

A. Stretching vibrations — bond length changes (atoms move along the bond axis).

1. **Symmetric stretching:** both bonds stretch in and out together (in phase).
2. **Asymmetric stretching:** one bond stretches while the other contracts (out of phase).

B. Bending vibrations — bond angle changes (atoms move out of the bond axis). Four types:

1 **Scissoring:** two atoms swing toward and away from each other in the plane — like scissors opening and closing.

2 **Rocking:** the group swings back and forth in the plane — like a rocking chair.

3 **Wagging:** the group swings back and forth out of the plane — like a dog wagging its tail up and down.

4 **Twisting:** the group twists out of the plane — one atom moves up, the other down.

Textual diagram — the four bending modes (methylene $-CH_2-$ group):

```
Scissoring (in-plane):          Rocking (in-plane):
    H   H                           H   H
     \ /                             \ /
      C       →  H–C–H angle          C      →  whole CH₂ rocks
     / \            opens/closes     / \         left–right
    H   H                           (rest of molecule)

Wagging (out-of-plane):         Twisting (out-of-plane):
    H                               H
     \                               \
      C  →  both H move               C  →  one H up,
     /      up/down together         /      one H down
    H                               H
```

Mnemonic for the four bends: "Scissors Rock, Wag, Twist" — **S**cissoring, **R**ocking, **W**agging, **T**wisting.

Which vibrations absorb IR? Only vibrations that cause a **change in dipole moment** are IR-active. (Symmetric stretching of CO_2, for example, causes no dipole change and is IR-inactive — but Raman-active.)

Key takeaway: Stretching needs more energy (higher wavenumber, ~4,000–1,500 cm^{-1}) than bending (lower wavenumber, ~1,500–400 cm^{-1}).

5.3 Hooke's Law — Vibrational Frequency Equation

A vibrating bond behaves like a spring, so it follows **Hooke's law** (the same law as a mass on a spring).

The equation (statement):

$$\bar{\nu} = (1 / 2\pi c) \times \sqrt{(k / \mu)}$$

where the frequency is expressed as **wavenumber $\bar{\nu}$ (cm^{-1})**.

Meaning of each term:

Term	Name	Meaning
$\bar{\nu}$	Wavenumber of vibration	The position of the absorption band (cm^{-1}) — what we read on the spectrum
c	Speed of light	3×10^{10} cm/s (constant)

Term	Name	Meaning
k	Force constant	Stiffness/strength of the bond (like spring stiffness); stronger bond → higher wavenumber
μ	Reduced mass	$\mu = (m_1 \cdot m_2)/(m_1 + m_2)$, where m_1, m_2 are the masses of the two atoms; lighter atoms → higher wavenumber

In words: Vibrational frequency **increases** with bond strength (k) and **decreases** with atomic mass (μ).

5.4 Factors Affecting Vibrations

Factor 1 — Reduced mass (μ): lighter atoms vibrate faster.

- **Example: C−H vs C−D.** Replacing hydrogen with heavier deuterium increases μ, so the C−D stretch appears at a **lower** wavenumber (~2,100 cm^{-1}) than C−H (~2,900 cm^{-1}).
- Rule: the bond with the lighter atom always absorbs at higher wavenumber.

Factor 2 — Force constant (k): stronger bonds vibrate faster.

- **Example: bond order.** C≡C (triple, ~2,100 cm^{-1}) > C=C (double, ~1,650 cm^{-1}) > C−C (single, ~1,000 cm^{-1}).
- Stronger spring = faster vibration = higher wavenumber.

CALCULATION BOX 6 — Wavenumber from wavelength

Question: An IR band appears at wavelength 5.88 μm. Calculate its wavenumber in cm^{-1}. (This is the carbonyl C=O region — check.)

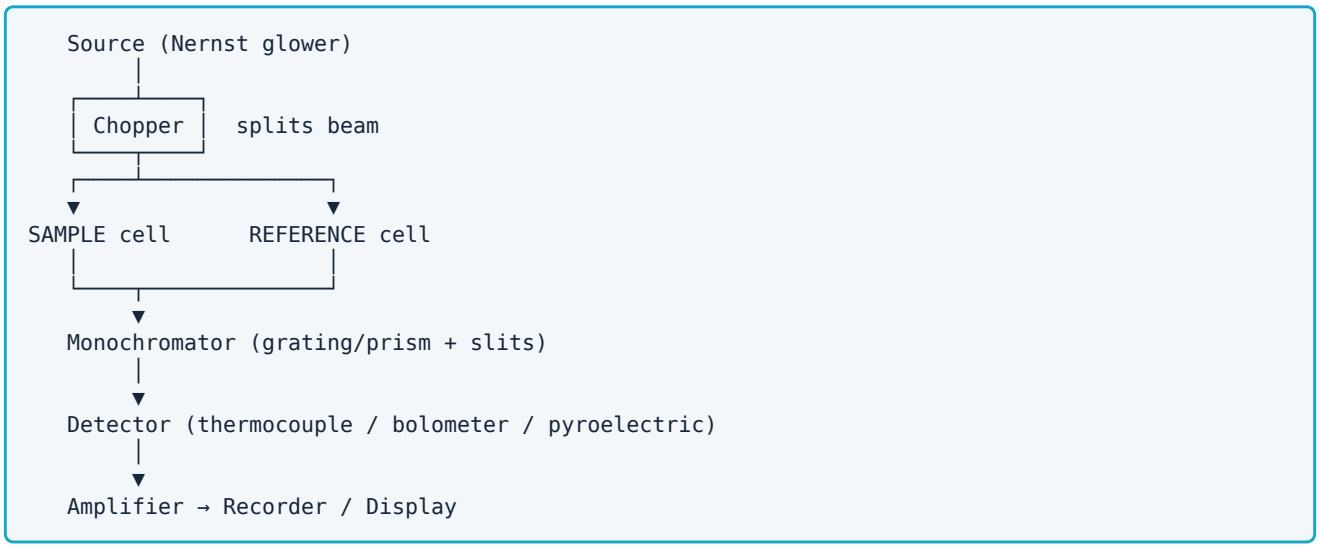
Solution: Step 1: Convert μm to cm: 5.88 μm = 5.88×10^{-4} cm. Step 2: $\bar{\nu} = 1/\lambda = 1 / (5.88 \times 10^{-4}$ cm) = 1,700 cm^{-1}. **Answer:** 1,700 cm^{-1} — yes, this is the characteristic C=O stretching region.

One-minute takeaway: "Light atoms, strong bonds → high wavenumber."

5.5 Instrumentation — Dispersive Infrared Spectrophotometer

A dispersive IR instrument is similar to a UV instrument, but built for IR: **source → sample → monochromator → detector → readout**.

Dispersive IR optical diagram (textual):



5.5.1 IR Source

- **Nernst glower:** a rod of rare-earth oxides (zirconium/yttrium oxides) heated electrically to ~1,500 °C; emits continuous IR radiation.
- **Globar:** a silicon carbide rod, heated similarly; sturdy and long-lived.
- (A tungsten lamp is NOT used — it is too weak in the IR region.)

5.5.2 Monochromator

- A **diffraction grating** (or prism of NaCl/KBr, which transmit IR) disperses the IR beam; slits select the wavenumber.
- In dispersive instruments the monochromator sits **after** the sample, so the sample is exposed to the full IR beam (unlike UV instruments).

5.5.3 IR Detectors (heat detectors — IR is detected as heat)

1. Thermocouple

- **Construction:** Two dissimilar metal wires joined at two junctions (a "thermopile" uses several pairs in series); one junction is blackened to absorb IR.
- **Working:** IR radiation heats the blackened junction → temperature difference between the junctions generates a small voltage (Seebeck effect) → voltage proportional to IR intensity.

2. Bolometer

- **Construction:** A thin strip of metal (e.g., platinum) or semiconductor whose electrical resistance changes with temperature.
- **Working:** IR radiation heats the strip → its resistance changes → measured as a change in current/voltage proportional to IR intensity.

3. Pyroelectric detector

- **Construction:** A crystal of pyroelectric material (e.g., deuterated triglycine sulphate, DTGS) placed between electrodes.
- **Working:** IR radiation changes the crystal's temperature → the crystal's electric polarisation changes → a measurable charge/current appears. Fast response; widely used in FTIR instruments.

Comparison table:

Detector	Principle	Speed	Use
Thermocouple	Voltage from temperature difference	Slow	Classic dispersive IR
Bolometer	Resistance change with temperature	Medium	Sensitive IR work
Pyroelectric	Polarisation change with temperature	Fast	FTIR instruments

5.6 Sample-Handling Techniques

IR samples must be prepared so that IR light can pass through. Four main techniques:

1. Nujol Mull

- **Method:** The solid sample is ground to a fine powder and mixed with a drop of **Nujol** (liquid paraffin, a mineral oil) to form a paste (mull); the paste is pressed between two NaCl plates.
- **Advantages:** Quick, simple, needs very little sample; no heat or pressure.
- **Limitations:** Nujol itself absorbs at ~2,900 cm^{-1} (C—H stretch) and ~1,450 cm^{-1}, hiding sample bands in those regions.

2. KBr Pellet (pressed disc)

- **Method:** ~1 mg of sample is ground with ~100 mg of dry **potassium bromide (KBr)** powder and pressed under high pressure into a transparent disc; KBr does not absorb IR.
- **Advantages:** No interfering bands (KBr is transparent in mid-IR); excellent spectra; standard pharmacopoeial method.
- **Limitations:** KBr absorbs moisture (must be dry); high pressure can change some crystal forms (polymorphs).

3. Thin Film (neat liquid)

- **Method:** A drop of the **pure liquid** sample is pressed between two NaCl plates to form a thin film.
- **Advantages:** Simplest for liquids; no solvent interference.
- **Limitations:** Only for liquids; film thickness hard to reproduce (not good for quantitation).

4. ATR (Attenuated Total Reflectance)

- **Method:** The sample (solid or liquid) is simply pressed against a crystal (diamond/ZnSe); the IR beam reflects inside the crystal and probes only the surface layer of the sample.
- **Advantages:** No sample preparation at all; non-destructive; works for solids, liquids, pastes.
- **Limitations:** Only the surface is measured (may not represent the bulk); weaker bands than transmission methods.

Comparison table:

Technique	Sample type	Preparation	Key limitation
Nujol mull	Solid	Grind + mineral oil paste	Nujol bands at 2,900 & 1,450 cm^{-1}
KBr pellet	Solid	Grind + press with KBr	Moisture; pressure effects
Thin film	Liquid	Drop between NaCl plates	Liquids only; thickness varies
ATR	Solid/liquid	Press on crystal	Surface-only measurement

Common mistake: Using glass or plastic plates for IR cells — glass absorbs IR strongly. IR cells and plates are made of **NaCl or KBr** (transparent to IR but water-soluble — keep them dry!).

5.7 FTIR — Fourier Transform Infrared Spectroscopy

Definition: FTIR is the modern form of IR spectroscopy in which all IR frequencies are measured simultaneously using an interferometer, and the spectrum is obtained by a mathematical operation called the Fourier transform.

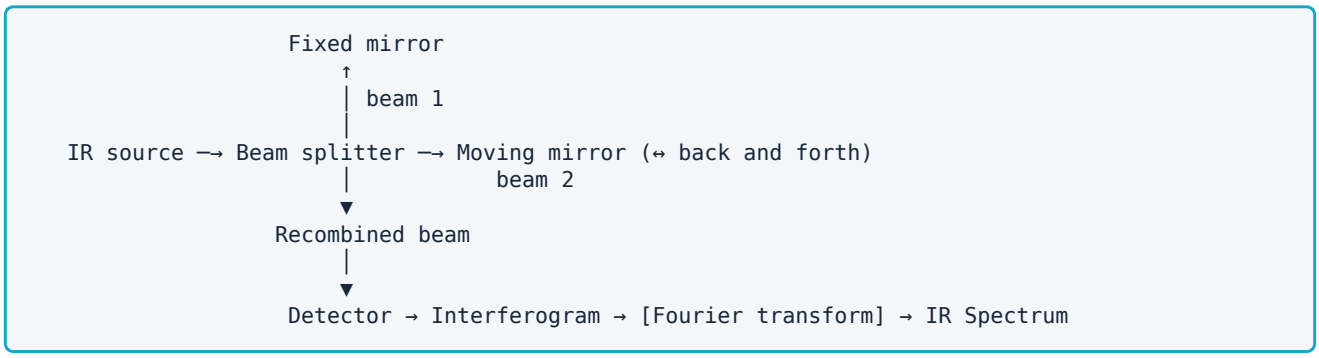
Principle — The Michelson Interferometer

Construction: A beam splitter, one fixed mirror, one moving mirror, and a detector.

Working (step by step):

1. The IR beam hits the **beam splitter** — half goes to the fixed mirror, half to the moving mirror.
2. Both beams reflect back and **recombine** at the beam splitter.
3. Because the moving mirror changes the path length, the two beams interfere — sometimes constructively (bright), sometimes destructively (dark).
4. The detector records intensity vs mirror position — this wavy signal is the **interferogram**.
5. A computer applies the **Fourier transform** (a mathematical operation that converts the time/position-domain signal into a frequency-domain spectrum) to get the normal IR spectrum (absorbance vs wavenumber).

Michelson interferometer diagram (textual):



Definition — Interferogram: The raw signal from an FTIR instrument — a plot of detector intensity versus mirror position (not versus wavenumber).

Definition — Fourier transform: The mathematical calculation that converts the interferogram into the familiar IR spectrum.

Advantages of FTIR (explained simply)

1. Fellgett advantage (multiplex advantage)

- In dispersive IR, each wavenumber is measured one at a time. In FTIR, **all wavenumbers are measured at the same time**.
- Result: a complete spectrum in seconds instead of minutes, and averaging many fast scans improves the signal quality.

2. Jacquinot advantage (throughput advantage)

- FTIR has no narrow slits blocking the beam, so **much more light reaches the detector**.
- Result: stronger signal, better sensitivity — tiny samples can be analysed.

Other advantages: better wavenumber accuracy (laser-calibrated), no stray light problems, computerised data handling.

One-minute takeaway: Fellgett = all frequencies at once (fast); Jacquinot = more light through (sensitive).

India context: Almost all new IR instruments purchased by Indian pharma companies and colleges today are FTIR instruments — dispersive IR survives mainly in older laboratories and in textbooks.

5.8 Pharmaceutical Applications of IR Spectroscopy

1. **Identification of drugs (fingerprint region):** The region **1,500–400 cm^{-1}** is called the **fingerprint region** — the pattern of bands here is unique to each molecule, like a human fingerprint. The IP identifies drugs by matching the sample spectrum with a reference spectrum.
2. **Functional-group detection:** Characteristic bands identify groups — O—H (~3,400 cm^{-1}), C=O (~1,700 cm^{-1}), N—H (~3,300 cm^{-1}). Confirms the structure of a synthesised drug.
3. **Polymorphism detection:** Different crystal forms (polymorphs) of the same drug give **different IR spectra** — critical because polymorphs differ in solubility and bioavailability (e.g., chloramphenicol palmitate polymorphs).

4 **Purity checking:** Extra or missing bands reveal impurities or degradation products.

5 **Raw-material verification:** Every incoming batch of drug raw material is IR-checked before manufacturing (identity test in the IP).

6 **Quantitative analysis:** Less common than UV, but possible using Beer–Lambert law with a characteristic band.

Pharmacy relevance: If a supplier sends "paracetamol" powder that is actually something else, the IR fingerprint will not match — the batch is rejected before a single tablet is made. This one test protects patients.

Infographic Concept Box — Chapter 5 One-Page Map

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INFRARED SPECTROSCOPY (bond vibrations)
├─ Regions: Near (12,800–4,000) · MID (4,000–400, workhorse) · Far (400–50 cm⁻¹)
├─ Vibrations: STRETCH (symmetric/asymmetric, high ν̄) · BEND (scissor/rock/wag/twist, low ν̄)
├─ Count: 3n–6 (non-linear) · 3n–5 (linear)   |   IR-active needs dipole change
├─ Hooke's law: ν̄ = (1/2πc)√(k/μ)  →  light atoms + strong bonds = high ν̄
├─ Dispersive IR: Source (Nernst/Globar) → Sample → Monochromator → Detector → Readout
├─ Detectors: Thermocouple (voltage) · Bolometer (resistance) · Pyroelectric (polarisation)
├─ Sampling: Nujol mull · KBr pellet · Thin film · ATR
├─ FTIR: Michelson interferometer → Interferogram → Fourier transform → Spectrum
│   └─ Fellgett (all λ at once = fast) · Jacquinot (no slits = more light = sensitive)
└─ Pharma: fingerprint ID (1,500–400 cm⁻¹) · functional groups · polymorphism · purity
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Key Points — Chapter 5

1 IR spectroscopy measures bond vibrations; mid IR (4,000–400 cm^{-1}) is the main region.

2 Vibrations: stretching (symmetric, asymmetric) and bending (scissoring, rocking, wagging, twisting).

3 Number of vibrations: 3n−6 (non-linear), 3n−5 (linear).

4 Hooke's law: $\bar{\nu} = (1/2\pi c)\sqrt{(k/\mu)}$; lighter atoms and stronger bonds → higher wavenumber.

5 Only vibrations with a change in dipole moment are IR-active.

6 Dispersive IR: Nernst glower/Globar source → sample → grating monochromator → heat detector.

7 Detectors: thermocouple (voltage), bolometer (resistance), pyroelectric (polarisation).

8 Sample handling: Nujol mull, KBr pellet, thin film, ATR — each with advantages and limitations.

9 FTIR: Michelson interferometer produces an interferogram; Fourier transform gives the spectrum.

10 Fellgett advantage (multiplex — all frequencies at once); Jacquinot advantage (throughput — more light).

11 Fingerprint region (1,500–400 cm^{-1}) identifies drugs; IR detects polymorphism and checks purity.

Common Confusions — Chapter 5

1 **Dispersive IR vs FTIR:** dispersive measures one wavenumber at a time using a monochromator; FTIR measures all at once using an interferometer — they are not the same instrument.

2 **Interferogram vs spectrum:** the interferogram is intensity vs mirror position (raw wiggly signal); the spectrum is absorbance vs wavenumber (after Fourier transform).

3 **Scissoring vs rocking:** both are in-plane bends — scissoring changes the H–C–H angle (like scissors); rocking moves the whole $-CH_2-$ group side to side (like a rocking chair).

4 **C−H vs C−D:** heavier deuterium vibrates slower → lower wavenumber. Heavier always means lower.

Common Mistakes — Chapter 5

1 Using glass cells for IR — glass absorbs IR; use NaCl/KBr plates and keep them dry.

2 Forgetting that Nujol has its own bands (2,900 and 1,450 cm^{-1}) — don't mistake them for sample peaks.

3 Saying "all 3n−6 vibrations appear in the spectrum" — only IR-active ones (dipole change) appear.

4 Mixing up Fellgett and Jacquinot — Fellgett = multiplex (simultaneous), Jacquinot = throughput (more light).

Mnemonics — Chapter 5

- Bending modes: **"Scissors Rock, Wag, Twist"**
- FTIR advantages: **"Fellgett = Fast (all at once); Jacquinot = Just more light"**
- Vibration rule: **"Light atoms, strong bonds → high wavenumber"**
- Regions: **"Near, Mid, Far — Mid is the star"**

Exam Summary — Chapter 5

- IR regions: near (12,800–4,000), mid (4,000–400), far (400–50 cm^{-1}); mid IR is the analytical workhorse.
- Vibration modes: symmetric/asymmetric stretching; scissoring, rocking, wagging, twisting; 3n−6/3n−5 rule; dipole-change requirement.
- Hooke's law: $\bar{\nu} = (1/2\pi c)\sqrt{(k/\mu)}$ with term meanings; reduced mass and force constant effects (C−H vs C−D; single/double/triple bonds).
- Dispersive IR diagram: source (Nernst glower/Globar) → sample → monochromator → detector (thermocouple/bolometer/pyroelectric) → readout.
- Sample handling: Nujol mull, KBr pellet, thin film, ATR — advantages/limitations.
- FTIR: Michelson interferometer → interferogram → Fourier transform → spectrum; Fellgett and Jacquinot advantages.
- Applications: fingerprint identification (1,500–400 cm^{-1}), functional groups, polymorphism, purity, raw-material verification.

One-Minute Revision — Chapter 5

IR = bond vibrations. Mid IR 4,000–400 cm^{-1}. Bends: scissor, rock, wag, twist. $\bar{\nu} = (1/2\pi c)\sqrt{(k/\mu)}$. Light atom + strong bond = high $\bar{\nu}$. KBr pellet is the IP standard; ATR needs no preparation. FTIR: interferometer → interferogram → Fourier transform. Fellgett = fast, Jacquinot = sensitive. Fingerprint region 1,500–400 cm^{-1} identifies drugs.

Exam Questions — Chapter 5

Short answers (2–5 marks):

1 Name the three IR regions with ranges. Which is most useful and why?

2 State the 3n−6 / 3n−5 rule and calculate the number of vibrations in H_2O.

3 State Hooke's-law equation for vibrational frequency and explain each term.

4 Why does C−D absorb at a lower wavenumber than C−H?

5 Name the four bending vibrations with a one-line description of each.

6 What is an interferogram?

7 State the Fellgett and Jacquinot advantages in one line each.

8 What is the fingerprint region? Why is it called that?

Long answers (10–15 marks):

1 Describe the fundamental modes of vibration in polyatomic molecules with diagrams. Derive the number of vibrations using the degrees-of-freedom concept.

2 Draw a labelled optical diagram of a dispersive IR spectrophotometer and explain the function of the source, monochromator and the three detectors.

3 Compare the four IR sample-handling techniques (Nujol mull, KBr pellet, thin film, ATR) with advantages and limitations of each.

4 Explain the principle of FTIR with the Michelson interferometer diagram. What is an interferogram, and what are the Fellgett and Jacquinot advantages?

5 Discuss the pharmaceutical applications of IR spectroscopy with emphasis on drug identification, polymorphism and purity checking.

MCQs (answer key at end):

1 The mid-IR region is: (a) 12,800–4,000 cm^{-1} (b) 4,000–400 cm^{-1} (c) 400–50 cm^{-1} (d) 200–800 nm → **(b)**

2 For a non-linear molecule with n atoms, the number of fundamental vibrations is: (a) 3n (b) 3n−5 (c) 3n−6 (d) 2n → **(c)**

3 According to Hooke's law, increasing the force constant: (a) decreases wavenumber (b) increases wavenumber (c) has no effect (d) doubles the mass → **(b)**

4 Which detector works on change of electrical resistance with temperature? (a) thermocouple (b) bolometer (c) pyroelectric (d) photomultiplier → **(b)**

5 The Fellgett advantage is also called the: (a) throughput advantage (b) multiplex advantage (c) resolution advantage (d) sensitivity advantage → **(b)**

6 KBr is used in IR sample preparation because it: (a) absorbs IR strongly (b) is transparent in the mid-IR (c) reacts with the sample (d) is coloured → **(b)**

7 The fingerprint region is: (a) 4,000–2,500 cm^{-1} (b) 2,500–1,500 cm^{-1} (c) 1,500–400 cm^{-1} (d) 400–50 cm^{-1} → **(c)**

8 ATR sampling is advantageous because: (a) it needs no sample preparation (b) it uses glass cells (c) it measures only UV (d) it needs large samples → **(a)**

Viva questions:

1 Why can't we use glass plates for IR cells? (Glass absorbs IR; NaCl/KBr plates are used.)

2 Your KBr pellet looks cloudy — what went wrong? (Moisture in KBr; dry it before use.)

3 What does the Fourier transform do in FTIR? (Converts the interferogram into the spectrum.)

4 Two polymorphs of a drug — how does IR help? (Different crystal forms give different IR spectra.)

Application-based:

1 A new batch of ibuprofen raw material arrives at a formulation plant in Hyderabad. Outline the IR-based identity check you would perform. (Prepare KBr pellet, record 4,000–400 cm^{-1} spectrum, compare fingerprint region with IP reference spectrum; confirm characteristic bands.)

2 A QC chemist records an IR spectrum using a Nujol mull and sees strong bands at 2,900 and 1,450 cm^{-1} that are absent in the reference KBr spectrum. Explain. (These are Nujol's own C−H bands, not sample peaks.)

Practice numerical (unworked, answer given):

1 An O−H stretching band appears at 3,400 cm^{-1}. Calculate the corresponding wavelength in μm.
Answer: 2.94 μm

References — Chapter 5:

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