

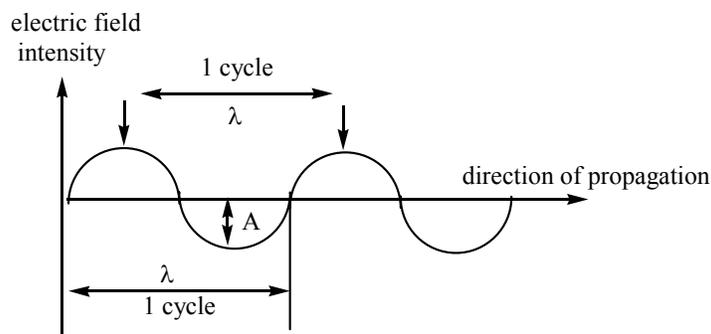
## Introduction

The molecular spectroscopy is the study of the interaction of electromagnetic waves and matter. The scattering of sun's rays by raindrops to produce a rainbow and appearance of a colorful spectrum when a narrow beam of sunlight is passed through a triangular glass prism are the simple examples where white light is separated into the visible spectrum of primary colors. This visible light is merely a part of the whole spectrum of electromagnetic radiation, extending from the radio waves to cosmic rays. All these apparently different forms of electromagnetic radiations travel at the same velocity but characteristically differ from each other in terms of frequencies and wavelength (Table 1).

**Table 1: The electromagnetic spectrum**

Radiation type	Wave length $\lambda$ , (Å)	Frequency $\nu = c / \lambda$ , (Hz)	Applications
radio	$10^{14}$	$3 \times 10^4$	
Nuclear magnetic resonance	$10^{12}$	$3 \times 10^6$	
Television	$10^{10}$	$3 \times 10^8$	Spin orientation
Radar	$10^8$	$3 \times 10^{10}$	
Microwave	$10^7$	$3 \times 10^{11}$	Rotational
Far infrared	$10^6$	$3 \times 10^{12}$	Vibrational
Near infrared	$10^4$	$3 \times 10^{14}$	
Visible	$8 \times 10^3$ - $4 \times 10^3$	$3.7 \times 10^{14}$ - $7.5 \times 10^{14}$	
Ultraviolet	$3 \times 10^3$	$1 \times 10^{15}$	Electronic
X-rays	1	$3 \times 10^{18}$	
Gamma rays	$10^{-2}$	$3 \times 10^{20}$	Nuclear transitions
Cosmic rays	$10^{-4}$	$3 \times 10^{22}$	

The propagation of these radiations involves both electric and magnetic forces which give rise to their common class name electromagnetic radiation. In spectroscopy, only the effects associated with electric component of electromagnetic wave are important. Therefore, the light wave traveling through space is represented by a sinusoidal trace (figure 1). In this diagram  $\lambda$  is the wavelength and distance A is known as the maximum amplitude of the wave. Although a wave is frequently characterized in terms of its wavelength  $\lambda$ , often the terms such as wavenumber ( $\bar{\nu}$ ), frequency ( $\nu$ ), cycles per second (cps) or hertz (Hz) are also used.



**Figure 1:** Wave like propagation of light ( $\lambda$  = wavelength ,  $A$  = amplitude)

The unit commonly used to describe the wavelength is centimeters (cm), the different units are used to express the wavelengths in different parts of the electromagnetic spectrum. For example, in the ultraviolet and visible region, the units use are angstrom ( $\text{\AA}$ ) and nanometer (nm). In the infrared region, the commonly used unit is wavenumber ( $\bar{\nu}$ ), which gives the number of waves per centimeter. Thus

$$1 \text{ cm} = 10^7 \text{ nm} = 10^8 \text{ \AA}$$

$$1 \text{ \AA} = 10^{-1} \text{ nm} = 10^{-8} \text{ cm}$$

The four quantities wavelength, wavenumber, frequency and velocity can be related to each other by following relationships

$$\text{Wavelength } (\lambda) = 1 / \bar{\nu} = c / \nu$$

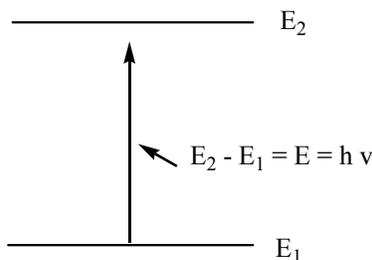
$$\text{Wave-number } (\bar{\nu}) = 1 / \lambda = \nu / c$$

$$\text{Frequency } (\nu) = c / \lambda = c \bar{\nu}$$

$$\text{Velocity } (c) = \nu \lambda = \nu / \bar{\nu}$$

### Absorption of Different Electromagnetic radiations by Organic Molecules

In absorption spectroscopy, though the mechanism of absorption of energy is different in the ultraviolet, infrared and nuclear magnetic resonance regions, the fundamental process is the absorption of a discrete amount of energy. The energy required for the transition from a state of lower energy ( $E_1$ ) to state of higher energy ( $E_2$ ) is exactly equivalent to the energy of electromagnetic radiation that causes transition.



**Figure 2:** Energy transition for the absorption of any electromagnetic radiation

$$\therefore E_1 - E_2 = E = h\nu = hc / \lambda$$

Where E is energy of electromagnetic radiation being absorbed, h is the universal Planck's constant,  $6.624 \times 10^{-27}$  erg sec and  $\nu$  is the frequency of incident light in cycles per second (cps or hertz, Hz), c is velocity of light  $2.998 \times 10^{10}$  cm s<sup>-1</sup> and  $\lambda$  = wavelength (cm)

Therefore, higher is the frequency, higher would be the energy and longer is the wavelength, lower would be the energy. As we move from cosmic radiations to ultraviolet region to infrared region and then radio frequencies, we are gradually moving to regions of lower energies.

A molecule can only absorb a particular frequency, if there exists within the molecule an energy transition of magnitude  $E = h\nu$

Although almost all parts of electromagnetic spectrum are used for understanding the matter, in organic chemistry we are mainly concerned with energy absorption from only ultraviolet and visible, infrared, microwave and radiofrequency regions.

**Ultraviolet – visible spectroscopy** ( $\lambda$  200 - 800 nm) studies the changes in electronic energy levels within the molecule arising due to transfer of electrons from  $\pi$ - or non-bonding orbitals. It commonly provides the knowledge about  $\pi$ -electron systems, conjugated unsaturations, aromatic compounds and conjugated non-bonding electron systems etc.

**Infrared spectroscopy** ( $\bar{\nu}$  400-4000 cm<sup>-1</sup>) studies the changes in the vibrational and rotation movements of the molecules. It is commonly used to show the presence or absence of functional groups which have specific vibration frequencies viz. C=O, NH<sub>2</sub>, OH, CH, C-O etc.

**Nuclear magnetic resonance** (radiofrequency  $\nu$  60-600 MHz) provides the information about changes in magnetic properties of certain atomic nuclei. <sup>1</sup>H and <sup>13</sup>C are the most commonly studied nuclei for their different environments and provide different signals for magnetically non-equivalent nuclei of the same atom present in the same molecule.

In the present chapter, UV-Vis and Infrared spectroscopy have been discussed.

## Ultraviolet and Visible Spectroscopy

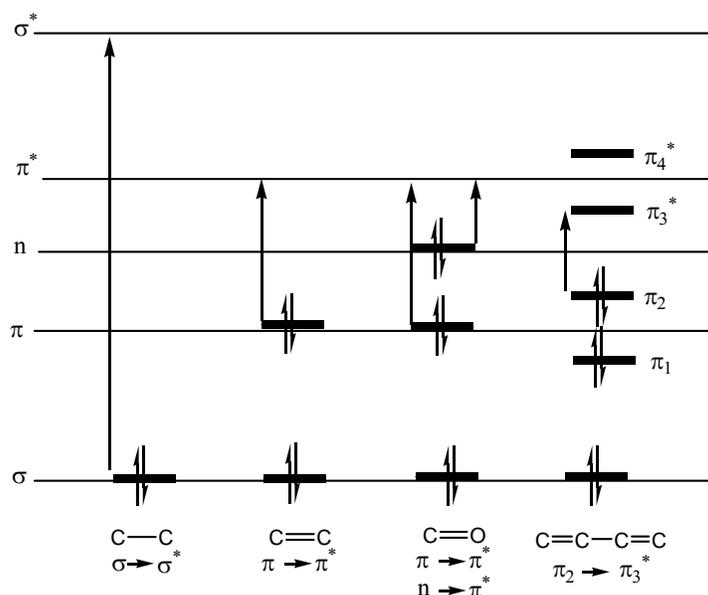
This absorption spectroscopy uses electromagnetic radiations between 190 nm to 800 nm and is divided into the ultraviolet (UV, 190-400 nm) and visible (VIS, 400-800 nm) regions. Since the absorption of ultraviolet or visible radiation by a molecule leads transition among electronic energy levels of the molecule, it is also often called as electronic spectroscopy. The information provided by this spectroscopy when combined with the information provided by NMR and IR spectral data leads to valuable structural proposals.

### Nature of Electronic Transitions

The total energy of a molecule is the sum of its electronic, its vibrational energy and its rotational energy. Energy absorbed in the UV region produces changes in the electronic energy of the molecule. As a molecule absorbs energy, an electron is promoted from an occupied molecular

orbital (usually a non-bonding  $n$  or bonding  $\pi$  orbital) to an unoccupied molecular orbital (an antibonding  $\pi^*$  or  $\sigma^*$  orbital) of greater potential energy (figure 3). For most molecules, the lowest-energy occupied molecular orbitals are  $\sigma$  orbitals, which correspond to  $\sigma$  bonds. The  $\pi$  orbitals lie at relatively higher energy levels than  $\sigma$  orbitals and the non-bonding orbitals that hold unshared pairs of electrons lie even at higher energies. The antibonding orbitals ( $\pi^*$  and  $\sigma^*$ ) are orbitals of highest energy. The relative potential energies of these orbitals and various possible transitions have been depicted in figure 3.

The saturated aliphatic hydrocarbons (alkanes) exhibit only  $\sigma \rightarrow \sigma^*$  transitions but depending on the functional groups the organic molecules may undergo several possible transitions which can be placed in the increasing order of their energies viz.  $n \rightarrow \pi^* < n \rightarrow \sigma^* < \pi \rightarrow \pi^* < \sigma \rightarrow \pi^* < \sigma \rightarrow \sigma^*$ . Since all these transitions require fixed amount of energy (quantized), an ultraviolet or visible spectrum of a compound would consist of one or more well defined peaks, each corresponding to the transfer of an electron from one electronic level to another. If the differences between electronic energy levels of two electronic states are well defined i.e. if the nuclei of the two atoms of a diatomic molecule are held in fixed position, the peaks accordingly should be sharp. However, vibrations and rotations of nuclei occur constantly and as a result each electronic state in a molecule is associated with a large

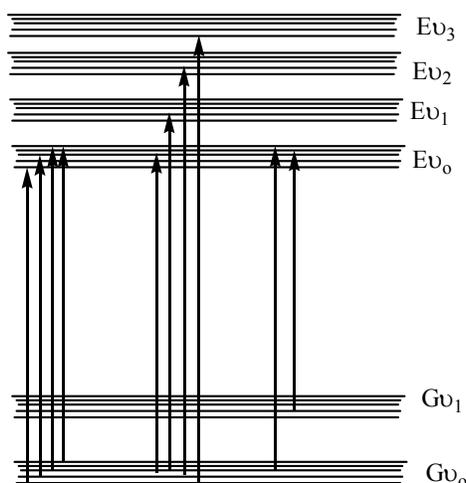


**Figure 3 :** Relative energies of orbitals most commonly involved in electronic spectroscopy of organic molecules

number of vibrational and rotational states. At room temperature, the molecules in the ground state will be in the zero vibrational level ( $G_{0,0}$ ). This is shown schematically in figure 4. The transition of an electron from one energy level to another is thus accompanied by simultaneous change in vibrational and rotational states and causes transitions between various vibrational and rotational levels of lower and higher energy electronic states. Therefore many radiations of closely placed frequencies are absorbed and a broad absorption band is obtained. When a molecule absorbs ultraviolet or visible light of a defined energy, an assumption is made that only one electron is excited from bonding orbital or non-bonding orbital to an anti-bonding orbital and

all other electrons remain unaffected. The excited state thus produced is formed in a very short time i.e. of the order of  $10^{-15}$  seconds. In accordance with Franck-Condon principle, during electronic excitation the atoms of the molecule do not move.

The most probable transition would appear to involve the promotion of one electron from the highest occupied molecular orbital (HOMO) to the lowest unoccupied molecular orbital (LUMO), but in many cases several transitions can be observed, giving several absorption bands in the spectrum. We can have a general view of the possible transitions prevailing in organic compounds.



**Figure 4 :** Energy level diagram showing excitation between different vibrational and rotational levels of two electronic states

Alkanes can only undergo  $\sigma \rightarrow \sigma^*$  transitions. These are high-energy transitions and involve very short wavelength ultraviolet light ( $< 150$  nm). These transitions usually fall outside the generally available measurable range of UV-visible spectrophotometers (200-1000 nm). The  $\sigma \rightarrow \sigma^*$  transitions of methane and ethane are at 122 and 135 nm, respectively. In alkenes amongst the available  $\sigma \rightarrow \sigma^*$  and  $\pi \rightarrow \pi^*$  transitions, the  $\pi \rightarrow \pi^*$  transitions are of lowest energy and absorb radiations between 170-190 nm.

In saturated aliphatic ketones the lowest energy transition involves the transfer of one electron of the nonbonding electrons of oxygen to the relatively low-lying  $\pi^*$  anti-bonding orbital. This  $n \rightarrow \pi^*$  transition is of lowest energy ( $\sim 280$  nm) but is of low intensity as it is symmetry forbidden. Two other available transitions are  $n \rightarrow \pi^*$  and  $\pi \rightarrow \pi^*$ . The most intense band for these compounds is always due to  $\pi \rightarrow \pi^*$  transition.

In conjugated dienes the  $\pi \rightarrow \pi^*$  orbitals of the two alkene groups combine to form new orbitals – two bonding orbitals named as  $\pi_1$  and  $\pi_2$  and two antibonding orbitals named as  $\pi_3^*$  and  $\pi_4^*$ . It is apparent that a new  $\pi \rightarrow \pi^*$  transition of low energy is available as a result of conjugation. Conjugated dienes as a result absorb at relatively longer wavelength than do isolated alkenes (see figure 6).

## Principles of Absorption Spectroscopy : Beer's and Lambert's Law

The greater the number of molecules that absorb light of a given wavelength, the greater the extent of light absorption and higher the peak intensity in absorption spectrum. If there are only a few molecules that absorb radiation, the total absorption of energy is less and consequently lower intensity peak is observed. This makes the basis of Beer-Lambert Law which states that the fraction of incident radiation absorbed is proportional to the number of absorbing molecules in its path.

When the radiation passes through a solution, the amount of light absorbed or transmitted is an exponential function of the molecular concentration of the solute and also a function of length of the path of radiation through the sample. Therefore,

$$\text{Log } I_0 / I = \epsilon c l$$

Where  $I_0$  = Intensity of the incident light (or the light intensity passing through a reference cell)

$I$  = Intensity of light transmitted through the sample solution

$c$  = concentration of the solute in  $\text{mol l}^{-1}$

$l$  = path length of the sample in cm

$\epsilon$  = molar absorptivity or the molar extinction coefficient of the substance whose light absorption is under investigation. It is a constant and is a characteristic of a given absorbing species (molecule or ion) in a particular solvent at a particular wavelength.  $\epsilon$  is numerically equal to the absorbance of a solution of unit molar concentration ( $c = 1$ ) in a cell of unit length ( $l = 1$ ) and its units are  $\text{liters.moles}^{-1} \cdot \text{cm}^{-1}$ . However, it is customary practice among organic chemists to omit the units.

The ratio  $I / I_0$  is known as transmittance  $T$  and the logarithm of the inverse ratio  $I_0 / I$  is known as the absorbance  $A$ .

Therefore

$$-\text{Log } I / I_0 = -\log T = \epsilon c l$$

and  $\text{Log } I_0 / I = A = \epsilon c l$

or  $A = \epsilon c l$

For presenting the absorption characteristics of a spectrum, the positions of peaks are reported as  $\lambda_{\text{max}}$  (in nm) values and the absorptivity is expressed in parenthesis.

## Spectral Measurements

The UV-Vis spectra are usually measured in very dilute solutions and the most important criterion in the choice of solvent is that the solvent must be transparent within the wavelength range being examined. **Table 2** lists some common solvents with their lower wavelength cut off limits. Below these limits, the solvents show excessive absorbance and should not be used to determine UV spectrum of a sample.

**Table 2 : Common solvents with their cut-off limits.**

S. No.	Solvent	Cut-off wavelength (nm)
1	Acetonitrile	190
2	Water	191
3	Cyclohexane	195
4	Hexane	201
5	Methanol	203
6	95% ethanol	304
7	1,4-dioxane	215
8	Ether	215
9	Dichloromethane	220
10	Chloroform	237
11	Carbon tetrachloride	257
12	Benzene	280

Of the solvents listed in **table 2**, water, 95% ethanol and hexane are the most commonly used solvents. Each is transparent in the region of UV-Vis spectrum. For preparing stock solutions, the sample is accurately weighed and made up to volume in volumetric flask. Aliquats are removed from this solution and appropriate dilutions are made to make solutions of desired concentration. For recording the spectrum 1 cm square quartz cell is commonly used. These require approx. 3 ml of solution. The quartz cell containing solution is placed in the path of light beam and spectrum is recorded by varying the wavelength of incident light.

### **Solvent Effects**

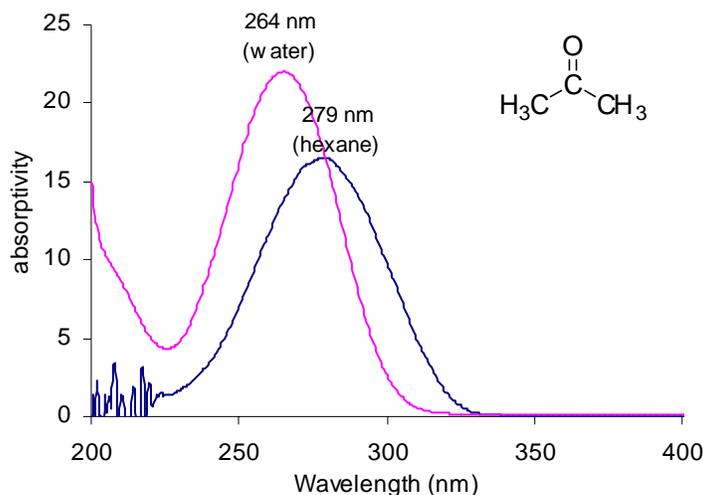
Highly pure, non-polar solvents such as saturated hydrocarbons do not interact with solute molecules either in the ground or excited state and the absorption spectrum of a compound in these solvents is similar to the one in a pure gaseous state. However, polar solvents such as water, alcohols etc. may stabilize or destabilize the molecular orbitals of a molecule either in the ground state or in excited state and the spectrum of a compound in these solvents may significantly vary from the one recorded in a hydrocarbon solvent.

#### **(i) $\pi \rightarrow \pi^*$ Transitions**

In case of  $\pi \rightarrow \pi^*$  transitions, the excited states are more polar than the ground state and the dipole-dipole interactions with solvent molecules lower the energy of the excited state more than that of the ground state. Therefore a polar solvent decreases the energy of  $\pi \rightarrow \pi^*$  transition and absorption maximum appears ~10-20 nm red shifted in going from hexane to ethanol solvent.

## (ii) $n \rightarrow \pi^*$ Transitions

In case of  $n \rightarrow \pi^*$  transitions, the polar solvents form hydrogen bonds with the ground state of polar molecules more readily than with their excited states. Therefore, in polar solvents the energies of electronic transitions are increased. For example, the figure 5 shows that the absorption maximum of acetone in hexane appears at 279 nm which in water is shifted to 264 nm, with a blue shift of 15 nm.



**Figure 5** : UV-spectra of acetone in hexane and in water

## Some important terms and definitions

**(i) Chromophore:** The energy of radiation being absorbed during excitation of electrons from ground state to excited state primarily depends on the nuclei that hold the electrons together in a bond. The group of atoms containing electrons responsible for the absorption is called chromophore. Most of the simple un-conjugated chromophores give rise to high energy transitions of little use. Some of these transitions have been listed in [table 3](#).

**Table 3:** Absorption maxima of simple un-conjugated chromophores

Chromophore	Transition	$\epsilon_{\text{max}}$	$\lambda_{\text{max}}$ (nm)
<i><math>\sigma</math> – bonded electrons</i>			
(C-C, C-H etc.)	$\sigma \rightarrow \sigma^*$		~ 150
<i>Lone pair electrons</i>			
-O-, -N-, -S-	$n \rightarrow \sigma^*$	100-1000	~ 190
C=O, C=N	$n \rightarrow \pi^*$	15	~300
	$\pi \rightarrow \pi^*$	500	~190
R-C $\equiv$ N	$n \rightarrow \pi^*$	5	~ 170
R-COOH, RCONH <sub>2</sub> , RCOOR	$n \rightarrow \pi^*$	50	~ 210

### *$\pi$ -bonded electrons*

C=C	$\pi \rightarrow \pi^*$	14000	~ 190
C $\equiv$ C	$\pi \rightarrow \pi^*$	2000	~ 195
C=O	$\pi \rightarrow \pi^*$	500	~ 180

For example, alkanes contain only single bonds with only possible  $\sigma \rightarrow \sigma^*$  type electronic transitions. These transitions absorb radiations shorter than wavelengths that are experimentally accessible in usually available spectrophotometers. In saturated molecules with heteroatom bearing non-bonding pairs of electrons,  $n \rightarrow \sigma^*$  transitions become available. These are also high energy transitions. In unsaturated compounds,  $\pi \rightarrow \pi^*$  transitions become possible. Alkenes and alkynes absorb ~ 170 nm but the presence of substituents significantly affects their position. The carbonyl compounds and imines can also undergo  $n \rightarrow \pi^*$  transitions in addition to  $\pi \rightarrow \pi^*$ . Amongst these, the most studied transitions are  $n \rightarrow \pi^*$  as these absorb at relatively longer wavelength 280-300 nm. These are low intensity ( $\epsilon$  10-100) transitions.

**(ii) Auxochrome:** The substituents that themselves do not absorb ultraviolet radiations but their presence shifts the absorption maximum to longer wavelength are called auxochromes. The substituents like methyl, hydroxyl, alkoxy, halogen, amino group etc. are some examples of auxochromes.

**(iii) Bathochromic Shift or Red shift:** A shift of an absorption maximum towards longer wavelength or lower energy.

**(iv) Hypsochromic Shift or Blue Shift:** A shift of an absorption maximum towards shorter wavelength or higher energy.

**(v) Hypochromic Effect:** An effect that results in decreased absorption intensity.

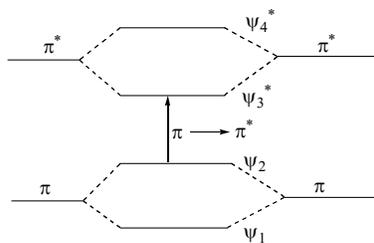
**(vi) Hyperchromic Effect:** An effect that results in increased absorption intensity.

## **Applications of Electronic Spectroscopy in Predicting Absorption Maxima of Organic Molecules**

### **1: Conjugated Dienes, Trienes and Polyenes**

The presence of conjugate double bond decreases the energy difference between HOMO and LUMO of resulting diene. The [figure 6](#) shows the change in energy of MO on conjugation. As a result, the radiations of longer wavelength are absorbed. The conjugation not only results in bathochromic shift (longer wavelength) but also increases the intensity of absorption. As the number of conjugated double bonds is increased, the gap between highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO) is progressively lowered. Therefore, the increase in size of the conjugated system gradually shifts the absorption maximum ( $\lambda_{\max}$ ) to longer wavelength and also increases the absorption. For example, ethylene absorbs at

175 nm ( $\epsilon = 1000$ ) and the conjugation in butadiene gives a strong absorption at longer wavelength at 230 nm and with higher intensity ( $\epsilon = >1000$ ).



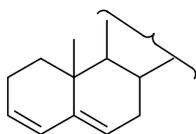
**Figure 6 :** Effect of conjugation on orbitals' energy and respective  $\pi$ - $\pi^*$  transition.

The presence of alkyl substituents on double bond also produces bathochromic shift and hyperchromic effect. These effects are additive in dienes and up to some extent in trienes. The open chain dienes can achieve *s-cis* or *s-trans* conformations and similarly diene system can be homoannular or heteroannular in cyclic systems. In 1941, Woodward suggested empirical rules for predicting the absorption of open chain and six-membered ring dienes which have been later on extended to large number of dienes and trienes (**Table 4**).

**Table 4: Empirical Rules for Diene and Triene absorptions**

Parent open chain or heteroannular diene	214 nm
Homoannular diene	253 nm
Increments for	
(a) each alkyl substituent or ring residue	5 nm
(b) double bond extending conjugation	30 nm
(c) exocyclic double bond	5 nm
(d) lone pair conjugation	
(i) O-C(=O)-R	0 nm
(ii) O-alkyl	6 nm
(iii) S-alkyl	30 nm
(iv) -Cl, -Br	5 nm
(v) NR <sub>2</sub>	60 nm

For example, here the absorption maxima for dienes **1** and **2** have been calculated according to Woodward rules. The comparison of calculated  $\lambda_{\max}$  values with observed  $\lambda_{\max}$  values highlights the importance of these rules.



1

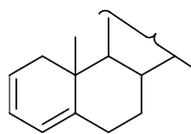
Parent value = 214 nm

Three ring residues (3 x 5) = 15

Exocyclic double bond = 5 nm

Total = 234 nm

Observed value = 235 nm



2

Parent value = 253 nm

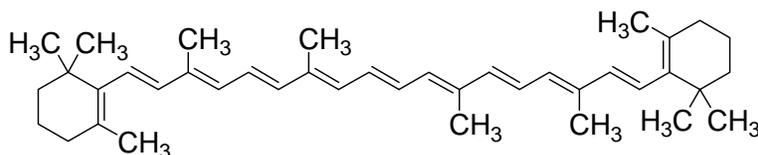
Three substituents = 15 nm

Ring residue = 5

Total = 273

Observed value = 275 nm

As the number of double bonds in conjugation increases, the bathochromic (towards longer wavelength) shift in lowest energy absorption maxima is observed. The increase in conjugation gradually shifts the maxima to visible region (> 400 nm) and imparts colour to the sample. **Table 5** shows the  $\lambda_{\max}$  shift in  $\text{Me}(\text{CH}=\text{CH})_n\text{Me}$  with increasing number of conjugated double bonds.  $\beta$ -Carotene (**figure 7**) responsible for red color in carrots is a typical example of polyene with 11 conjugated double bonds and exhibits  $\lambda_{\max}$  at 445 nm.



**Figure 7:** Structure of  $\beta$ -carotene

**Table 5:**  $\lambda_{\max}$  values for  $\text{Me}(\text{CH}=\text{CH})_n\text{Me}$

n	Wavelength (nm)
3	275
4	310
5	342
6	380
7	407

## 2: Carbonyl Compounds

Carbonyl compounds have two principal UV radiations, the allowed  $\pi \rightarrow \pi^*$  transitions and the forbidden  $n \rightarrow \pi^*$  transitions. In amides, acids, esters or acid halides, the substituents viz.  $\text{NR}_2$ , OH, OR, or  $-\text{X}$  on carbonyl group show pronounced hypsochromic effect on the  $n \rightarrow \pi^*$  transitions. The hypsochromic effect is due to inductive effect of nitrogen, oxygen or halogen atoms. The heteroatom withdraws electrons from carbonyl carbon and makes carbonyl oxygen lone pair of electrons more stabilized due to its involvement in increasing  $\text{C}=\text{O}$  bond order. As a result, the  $n \rightarrow \pi^*$  transition of these compounds is shifted to 200-215 nm range relative to 270 nm in aldehydes and ketones. Conjugation of the carbonyl group with double bond shifts both  $n \rightarrow \pi^*$  and  $\pi \rightarrow \pi^*$  transitions to longer wavelengths. The effect on  $\pi \rightarrow \pi^*$  band is more pronounced. Woodward formulated rules to predict the position of an absorption maximum in an unknown enone. These rules have been summarized in [table 6](#).

**Table 6: Empirical Rules for  $\alpha$ ,  $\beta$ -unsaturated ketones and aldehydes absorption maxima.**

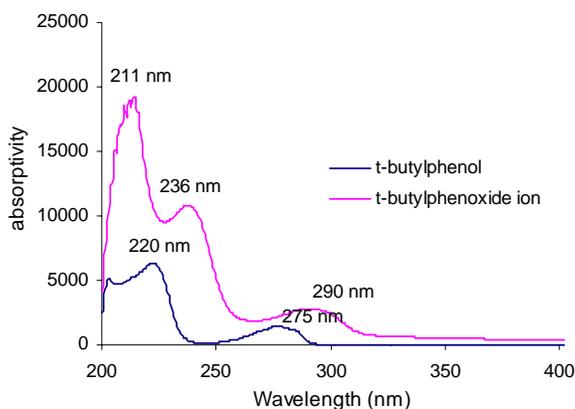
Parent acyclic ketone or six-membered		215 nm
Parent $\alpha$ , $\beta$ -unsaturated pentanone		202 nm
Parent $\alpha$ , $\beta$ -unsaturated aldehyde		207 nm
<i>Increments for</i>		
(a) a double bond extending conjugation		30 nm
(b) each alkyl group or ring residue	$\alpha$	10 nm
	$\beta$	12 nm
	$\gamma$ or higher	18 nm
<i>(c) auxochrome</i>		
(i) $-\text{OH}$	$\alpha$	35 nm
	$\beta$	30 nm
	$\delta$	50 nm
(ii) $\text{OCOR}$	$\alpha, \beta, \delta$	6 nm
(iii) $\text{OCH}_3$	$\alpha$	35 nm
	$\beta$	30 nm
	$\gamma$	17 nm
	$\delta$	31 nm
(iv) $\text{Cl}$	$\alpha$	15 nm
	$\beta$	12 nm
(v) $\text{Br}$	$\alpha$	25 nm
	$\beta$	30 nm
(vi) $\text{NR}_2$	$\beta$	95 nm
(d) exocyclic double bond		5 nm
(e) Homocyclic diene		30 nm

### 3. Aromatic Compounds

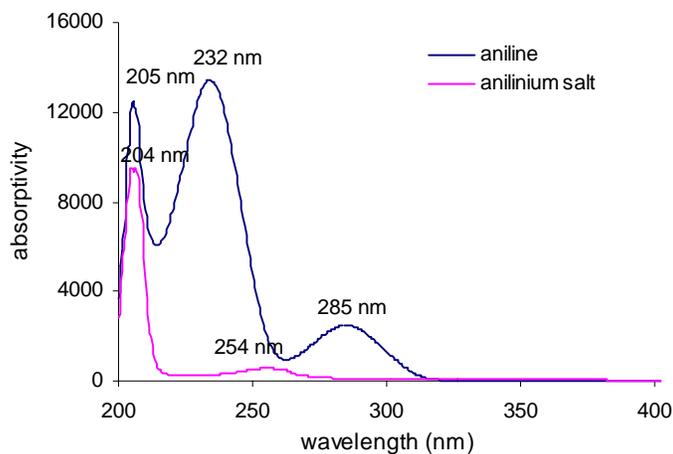
The simplest aromatic compound is benzene. It shows two primary bands at 184 ( $\epsilon = 47,000$ ) and 202 ( $\epsilon = 7400$ ) nm and a secondary fine structure band at 255 nm ( $\epsilon = 230$  in cyclohexane). Substituents on the benzene ring also cause bathochromic and hypsochromic shifts of various peaks. Unlike dienes and unsaturated ketones, the effects of various substituents on the benzene ring are not predictable. However, qualitative understanding of the effects of substituents on the characteristics of UV-Vis spectrum can be considered by classifying the substituents into electron-donating and electron-withdrawing groups.

**(i) Effect of Substituents with Unshared Electrons:** The non-bonding electrons increase the length of  $\pi$ -system through resonance and shift the primary and secondary absorption bands to longer wavelength. More is the availability of these non-bonding electrons, greater the shift will be. In addition, the presence of non-bonding electrons introduces the possibility of  $n \rightarrow \pi^*$  transitions. If non-bonding electron is excited into the extended  $\pi^*$  chromophore, the atom from which it is removed becomes electron-deficient and the  $\pi$ -system of aromatic ring becomes electron rich. This situation causes a separation of charge in the molecule and such excited state is called a charge-transfer or an electron-transfer excited state.

In going from benzene to *t*-butylphenol, the primary absorption band at 203.5 nm shifts to 220 nm and secondary absorption band at 254 nm shifts to 275 nm. Further, the increased availability of *n* electrons in negatively charged *t*-butylphenoxide ion shifts the primary band from 203.5 to 236 nm (a 32.5 nm shift) and secondary band shifts from 254 nm to 290 nm (a 36 nm shift) (Figure 8). Both bands show hyperchromic effect. On the other hand, in the case of anilinium cation, there are no *n* electrons for interaction and absorption properties are quite close to benzene. But in aniline, the primary band is shifted to 232 nm from 204 nm in anilinium cation and the secondary band is shifted to 285 nm from 254 nm (Figure 9).

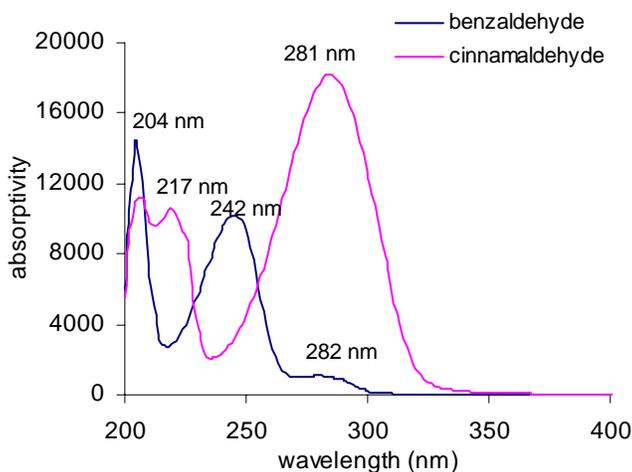


**Figure 8 :** UV-spectra of *t*-butyl phenol and *t*-butyl phenoxide in methanol



**Figure 9** : UV-spectra of aniline and anilinium salt in methanol

**(ii) Effect of  $\pi$  Conjugation:** Conjugation of the benzene ring also shifts the primary band at 203.5 nm more effectively to longer wavelength and secondary band at 254 nm is shifted to longer wavelength to lesser extent. In some cases, the primary band overtakes the secondary band. For example, benzoic acid shows primary band at 250 nm and secondary band at 273 nm, but cinnamic acid that has longer chromophore exhibits primary band at 273 nm and secondary band remains merged with it. Similarly, in benzaldehyde, the secondary band appears at 282 nm and primary band at 242 nm but in case of cinnamaldehyde, primary band appears at 281 nm and remains merged with secondary band (figure 10). The hyperchromic effect arising due to extended conjugation is also visible.

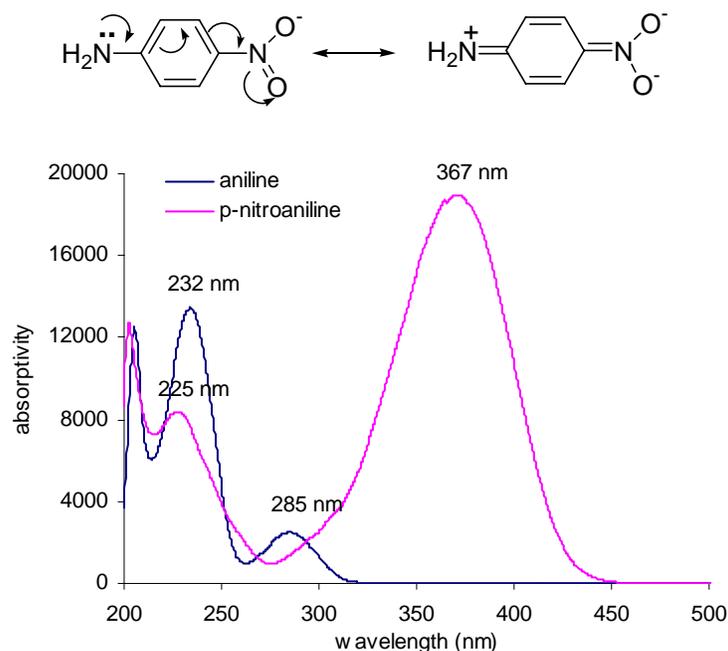


**Figure 10** : UV-spectra of benzaldehyde and cinnamaldehyde in methanol

**(iii) Effect of Electron-withdrawing and Electron-releasing Groups:** Electron-withdrawing substituents viz.  $\text{NH}_3^+$ ,  $\text{SO}_2\text{NH}_2$ ,  $\text{CN}$ ,  $\text{COOH}$ ,  $\text{COCH}_3$ ,  $\text{CHO}$  and  $\text{NO}_2$  etc. have no effect on the position of secondary absorption band of benzene ring. But their conjugation effects with  $\pi$ -electrons of the aromatic ring are observed. Electron-donating groups such as  $-\text{CH}_3$ ,  $-\text{Cl}$ ,  $-\text{Br}$ ,  $-\text{OH}$ ,  $-\text{OCH}_3$ ,  $-\text{NH}_2$  etc increase both  $\lambda_{\text{max}}$  and  $\epsilon_{\text{max}}$  values of the secondary band.

In case of disubstituted benzene derivatives, it is essential to consider the effect of both the substituents.

In para-substituted benzenes, two possibilities exist. If both the groups are electron-donating then the observed spectrum is closer to monosubstituted benzene. The group with stronger effect determines the extent of shifting of primary band. If one group is electron-releasing and other is electron-withdrawing, the magnitude of red shift is greater compared to the effect of single substituent individually. This is attributed to the increased electron drift from electron-donating group to the electron-withdrawing group through  $\pi$ -bond of benzene ring. For example, aniline shows secondary band at 285 nm which due to presence of electron-withdrawing *p*-nitro substituent is shifted to 367 nm with a significant increase in absorptivity (figure 11).



**Figure 11** : UV-spectra of aniline and *p*-nitroaniline in methanol

If two groups of a disubstituted benzene derivative are placed ortho- or meta- to each other, the combined effect of two substituents is observed. In case of substituted benzoyl derivatives, an empirical correction of structure with observed position of the primary absorption band has been developed. In the absence of steric hindrance to co-planarity, the calculated values are within  $\pm 5$  nm of the observed value.

**(iv) Polycyclic Aromatic Compounds:** In case of polycyclic aromatic hydrocarbons, due to extended conjugation, both primary and secondary bands are shifted to longer wavelength. These spectra are usually complicated but are characteristic of parent compound. The primary band at 184 nm in benzene shifts to 220 nm in case of naphthalene and 260 nm in case of anthracene.

Similarly, the structured secondary band which appears as broad band around 255 nm in benzene is shifted to 270 nm and 340 nm respectively in case of naphthalene and anthracene molecules.

### **Commercial Applications of UV and Visible Spectroscopy**

The UV-Vis spectroscopy has innumerable applications in the drugs and pharmaceutical industry.

Beer-Lambert law offers a valuable and simple method for quantitative analysis. In practice, a calibration curve is constructed by plotting absorbance vs. molar concentration and the concentration of unknown with 'X' absorbance is determined by finding the concentration corresponding to the measured absorbance on the calibration curve. The UV spectroscopy is used extensively in determining rate constants, equilibrium constants, acid-base dissociation constants etc for chemical reactions. The use of UV spectrometry in evaluation of enzymatic assays has become very common e.g. the activity of enzyme dehydrase is assayed by measuring the formation of ergosterol at 282 nm.

### **Infrared Absorption Spectroscopy**

The two atoms joined together by a chemical bond (may be single, double or triple bond), macroscopically can be composed as two balls joined by a spring. The application of a force like (i) stretching of one or both the balls (atoms) away from each other or closer to each other (ii) bending of one of the atoms either vertically or horizontally and then release of the force results in the vibrations on the two balls (atoms). These vibrations depend on the strength of the spring and also the mode (stretching or bending) in which the force is being applied.

Similarly, at ordinary temperatures, organic molecules are in a constant state of vibrations, each bond having its characteristic stretching and bending frequencies. When infrared light radiations between  $4000-400\text{ cm}^{-1}$  (the region most concerned to an organic chemist) are passed through a sample of an organic compound, some of these radiations are absorbed by the sample and are converted into energy of molecular vibrations. The other radiations which do not interact with the sample are transmitted through the sample without being absorbed. The plot of % transmittance against frequency is called the infrared spectrum of the sample or compound.

This study of vibrations of bonds between different atoms and varied multiplicities which depending on the electronegativity, masses of the atom and their geometry vibrate at different but specified frequencies; is called infrared spectroscopy. The presence of such characteristic vibrational bands in an infrared spectrum indicates the presence of these bonds in the sample under investigation.

### **Hooke's law and Absorption of radiations**

The band positions in the IR spectrum are presented in wave numbers ( $\bar{\nu}$ ) whose unit is the reciprocal centimeter ( $\text{cm}^{-1}$ ).  $\bar{\nu}$  is proportional to the energy of vibration.

$$\Delta E = h\nu = hc / \lambda = hc \bar{\nu}$$

Therefore, in principle, each absorption of radiation in the infrared region is quantized and should appear as sharp line. However, each vibrational transition within the molecule is