Spectrochemical analysis

UV-VIS spectroscopy

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2022-2023

outline

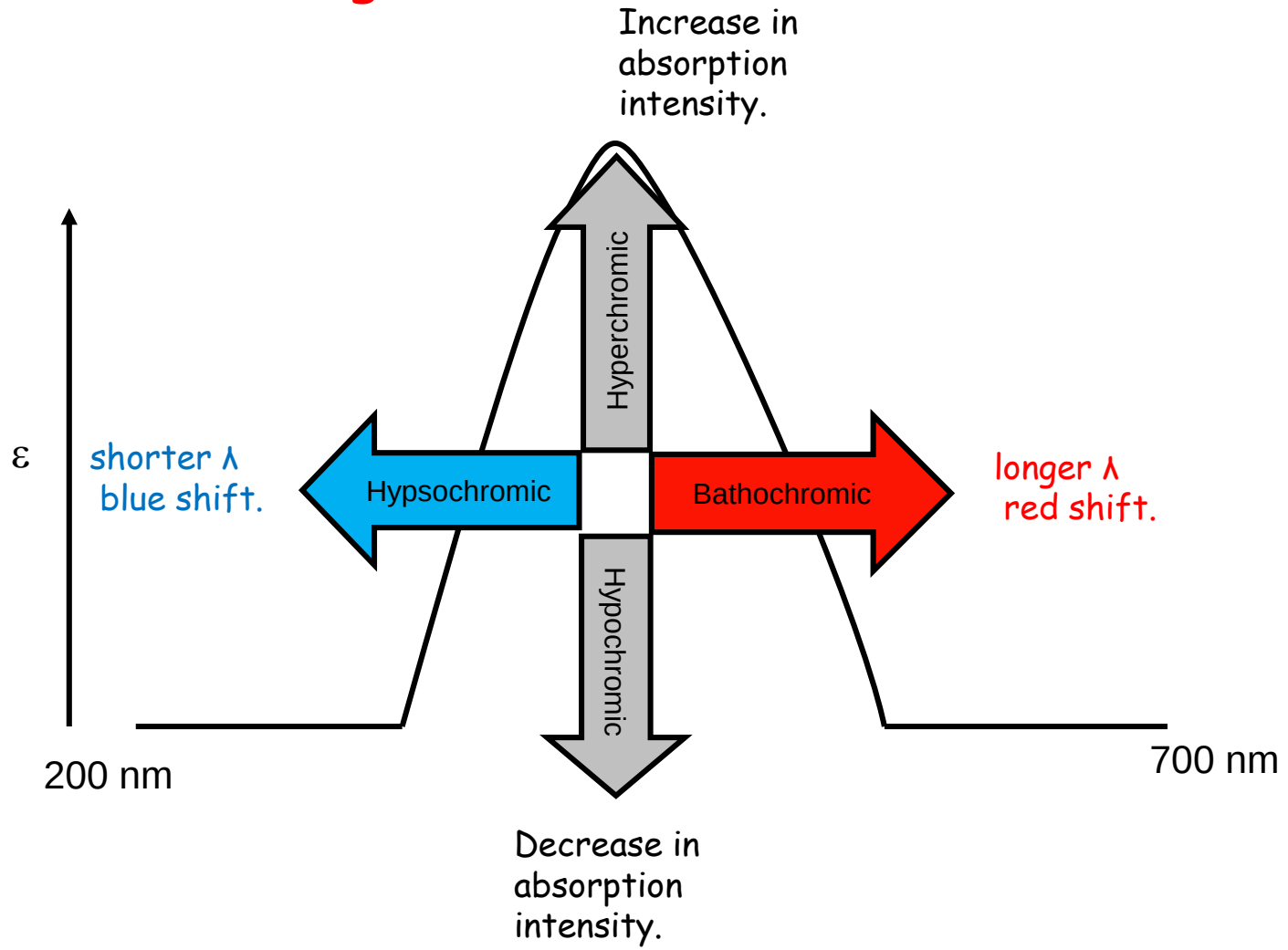
- Electromagnetic radiation (EMR)
- Wave Characteristics
- The particle nature of light
- Spectroscopy
- Absorption and Emission
- Beer-lambert's law
- Terms describing UV absorptions
- Molecular transitions
- Factors affecting UV absorptions
- Instrumentation
- Woodward –Fieser's rules

Terms describing UV absorptions

- **Chromophores:** Unsaturated organic functional groups that absorb in the ultraviolet or visible regions.

Group	Structure	λ_{max}
Carbonyl	> C = O	280
Azo	-N = N-	262
Nitro	-N=O	270
Thioketone	-C =S	330
Nitrite	-NO2	230
Conjugated Diene	-C=C-C=C-	233
Conjugated Triene	-C=C-C=C-C=C-	268
Conjugated Tetraene	-C=C-C=C-C=C-C=C-	315
Benzene		261

❖ **Auxochromes**: substituents with unshared pairs like OH, NH, SH ..., when attached to π chromophore they generally move the absorption max. to longer λ.



Uv-visible spectroscopy

❑ Why we use UV-Visible spectroscopy?

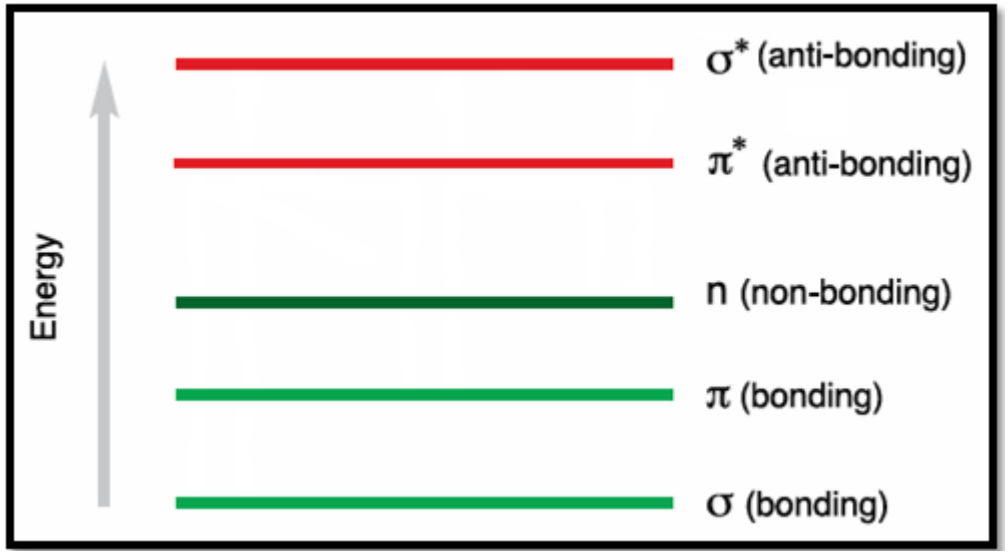
- ❑ Detection of functional groups
- ❑ Detection of impurities
- ❑ Qualitative and quantitative analysis
- ❑ Its helpful to show the relationship between different groups, and detect the conjugation of compounds.

- ❑ Types of electrons

σ electrons: in saturated compounds

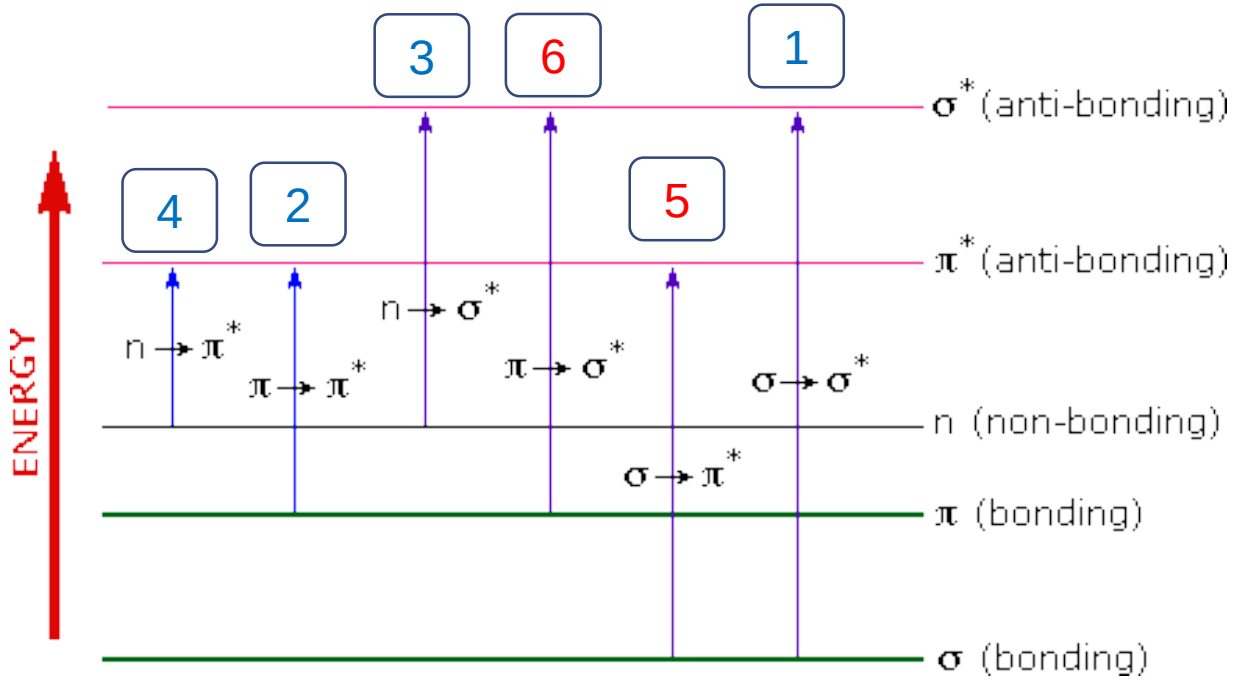
π electrons: in unsaturated compounds

n electrons: in non bonded electrons



Molecular transitions

- $\sigma \rightarrow \sigma^*$ transition
- $\pi \rightarrow \pi^*$ transition
- $n \rightarrow \sigma^*$ transition
- $n \rightarrow \pi^*$ transition
- $\sigma \rightarrow \pi^*$ transition
- $\pi \rightarrow \sigma^*$ transition



$n \rightarrow \pi^*$	$n \rightarrow \sigma^*$	$\pi \rightarrow \pi^*$	$\sigma \rightarrow \sigma^*$

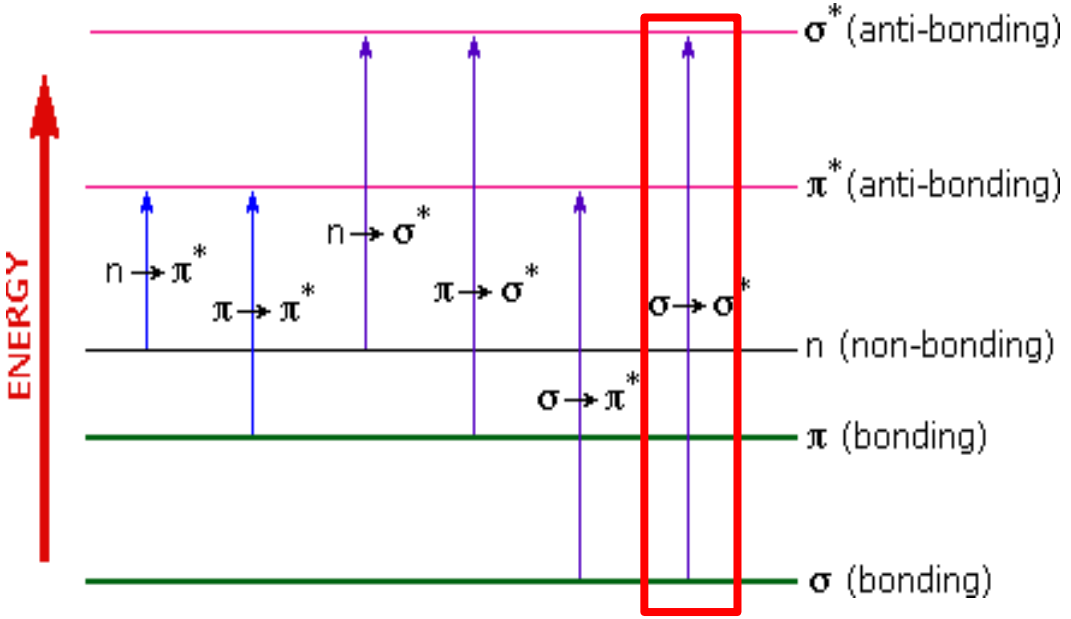
High energy (E)
Low wavelength (λ)

σ → σ* transition

- An electron in a bonding σ orbital is excited to the corresponding antibonding orbital. The energy required is large.

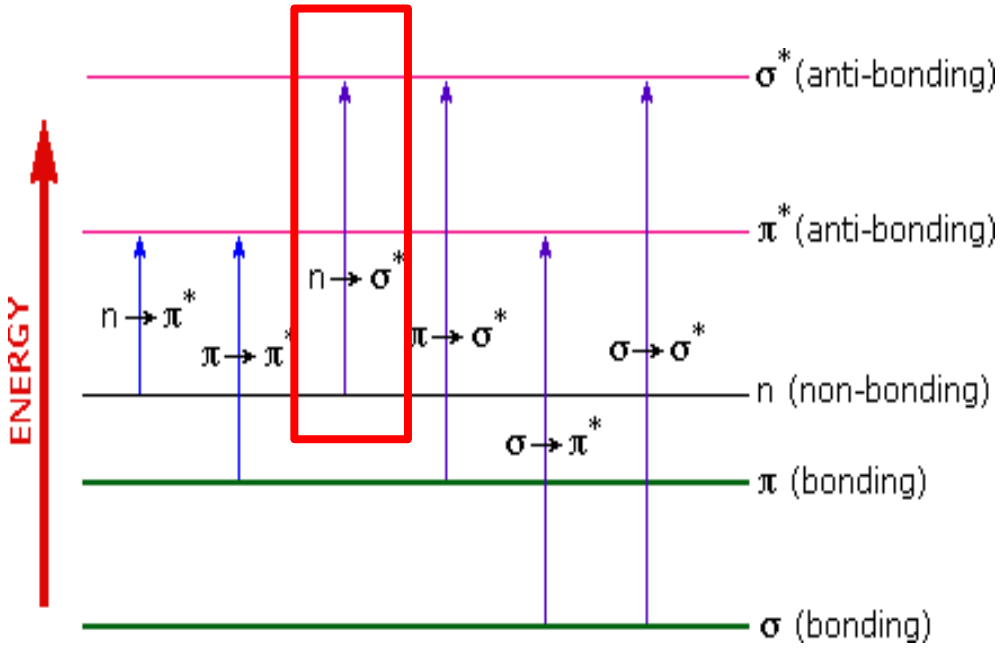
- For example, methane (CH_4) (which has only C-H bonds, and can only undergo σ → σ* transitions) shows an absorbance maximum at 125 nm.

- Absorption maxima due to σ → σ* transitions are not seen in typical UV-VIS spectra (200 - 700 nm)



$n \rightarrow \sigma^*$ transition

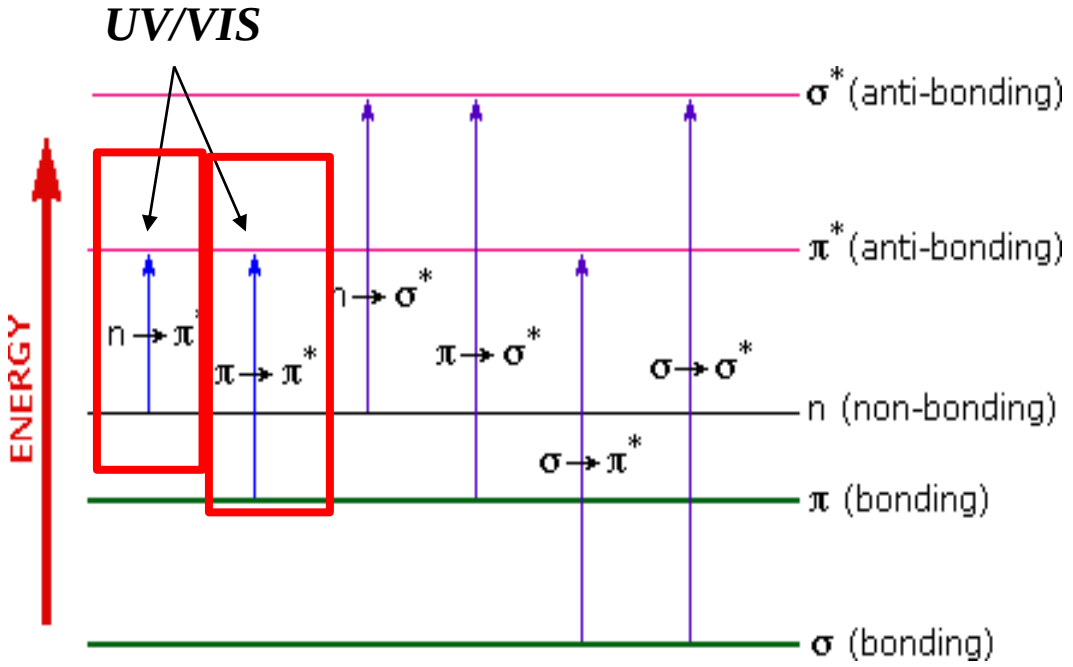
- Saturated compounds containing atoms with lone pairs (non-bonding electrons) are capable of $n \rightarrow \sigma^*$ transitions.
- These transitions usually need less energy than $\sigma \rightarrow \sigma^*$ transitions. They can be initiated by light whose wavelength is in the range 150 - 250 nm.
- The number of organic functional groups with $n \rightarrow \sigma^*$ peaks in the UV region is small.



n → π* transition

π → π* transition

- Most absorption spectroscopy of organic compounds is based on transitions of *n* or p electrons to the p* excited state.
- These transitions fall in an experimentally convenient region of the spectrum (200 - 700 nm).
- These transitions need an unsaturated group in the molecule to provide the p electrons.



- ***n*** → π* have low (∈) (10-100 L.mol^{-1}.cm^{-1})
- π → π* have high (∈) (1000-10000 L.mol^{-1}.cm^{-1})
- With highly polar solvent, (***n*** → π*) are shifted to lower (λ) (*blue shift*), due to unpaired electrons.
- With highly polar solvent, (π → π*) are shifted to higher (λ) (*red shift*), because of the attractive polarisation forces between solvent and absorbent.

$(\pi \rightarrow \pi^*)$ Transition is the most convenient and useful transition in UV-Vis Spectroscopy. Why?

π–π^* transition is the most frequently used transition for the following reasons:

- The ϵ for the π–π^* transition is high allowing sensitive determinations.
- The energy required is moderate, far less than dissociation energy.
- In presence of the most convenient solvent (water), the energy required for a π–π^* transition is usually smaller.

Chromophore	Excitation	λ_{max}, nm	Solvent
C=C	$\pi \rightarrow \pi^*$	171	hexane
C=O	$n \rightarrow \pi^*$ $\pi \rightarrow \pi^*$	290 180	hexane hexane
N=O	$n \rightarrow \pi^*$ $\pi \rightarrow \pi^*$	275 200	ethanol ethanol
C-X X=Br, I	$n \rightarrow \sigma^*$ $n \rightarrow \sigma^*$	205 255	hexane hexane

Factors influencing uv-vis absorption

- Type of solvent
- pH of the solution
- Temperature
- Concentration
- Conjugation

- This depends on the nature of the interaction of the particular solvent with the environment of the chromophore in the molecule under study.
- It is usually observed that ethanol solutions give absorption maxima at longer λ than hexane solutions.
- Changes in the polarity of the solvent can influence shifts to longer or shorter λ, by changing in the energy gap between these electronic states.

- **Non-polar solvents** (saturated hydrocarbons) **do not** interact with solute molecules either in the ground or excited state.

Factors influencing uv-vis absorption

- Type of solvent
- pH of the solution
- Temperature
- Concentration
- Conjugation

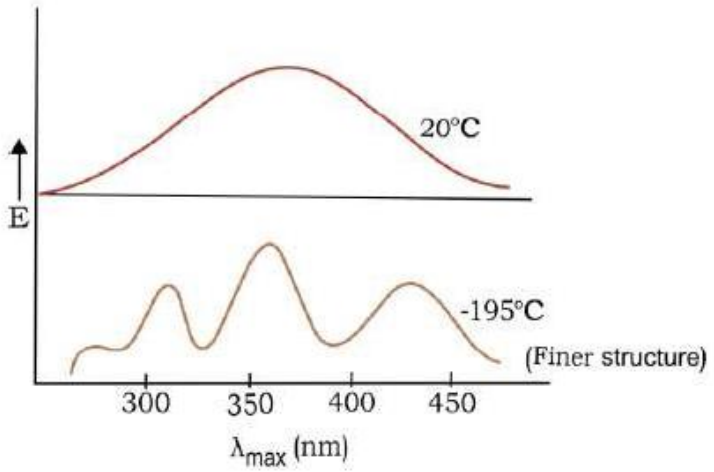
- The buffer though needs to be transparent over the wavelength range of the measurements. If the buffer absorbs radiation, absorbance readings attributed to the analyte may be higher than they should because the buffer and analyte absorptions will add together at each wavelength.
- If the optimum pH buffer solution is suitable with analyte, The absorbance spectra will show clear peak of the analyte, and vice versa.

Factors influencing uv-vis absorption

- Type of solvent
- pH of the solution
- Temperature
- Concentration
- Conjugation

- To get more accurate results, the spectrum needs to taken at a specified or constant temperature.

1. Band sharpness increases with decreasing temperature.
2. Position of absorption maximum does not move or moves very little towards the longer wavelength side, with decreasing temperature.
3. The total absorption intensity is approximately independent of the temperature.



Factors influencing uv-vis absorption

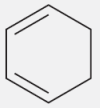
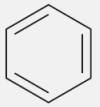
- Type of solvent
- pH of the solution
- Temperature
- Concentration
- Conjugation

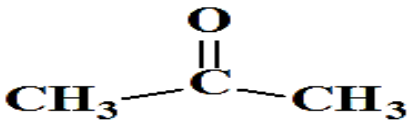
- Sample concentration is proportional to the intensity of the absorption.
- At high concentrations however, molecular interactions can take place causing changes to the position and shape of absorption bands.
- Such an outcome can affect the linearity of the relationship between sample concentration and absorbance.
- Higher the concentration of the analyte show higher absorbance they probably don't follow Beer-Lambert's law.
- Optimum concentration of analyte will be use , the absorbance will below then (0.8) .

Factors influencing uv-vis absorption

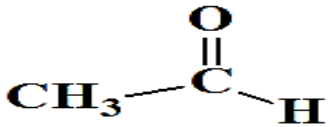
- Type of solvent
- pH of the solution
- Temperature
- Concentration
- Conjugation

- Wavelength of UV radiation that causes $\pi \rightarrow \pi^*$ excitation in a conjugated molecule ultimately depends on the nature of the conjugated system
- Degree of conjugation has a significant influence on the wavelength and molar absorptivity.

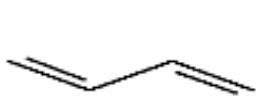
Name	Structure	λ_{max} (nm)
2-Methyl-1,3-butadiene	$H_2C{=}C(CH_3){-}CH{=}CH_2$	220
1,3-Cyclohexadiene		256
1,3,5-Hexatriene	$H_2C{=}CH{-}CH{=}CH{-}CH{=}CH_2$	258
1,3,5,7-Octatetraene	$H_2C{=}CH{-}CH{=}CH{-}CH{=}CH{-}CH{=}CH_2$	290
3-Buten-2-one	$H_2C{=}CH{-}C(=O){-}CH_3$	219
Benzene		203



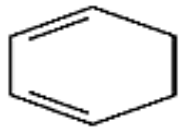
$\varepsilon = 19$



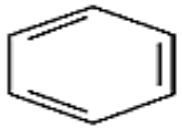
$\varepsilon = 12$



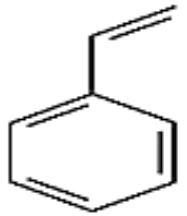
$\varepsilon = 20,900$



$\varepsilon = 4,580$



$\varepsilon = 8,000$



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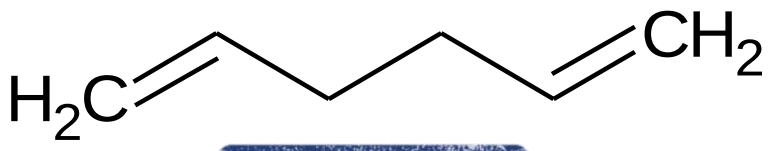
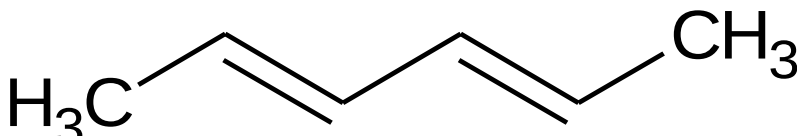
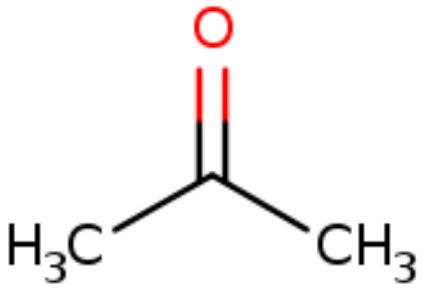
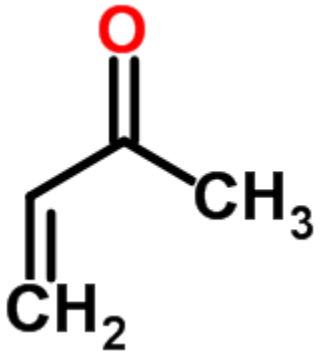


λ_{max}= 178 nm



λ_{max}= 227 nm





n → π*	λ_{max}= 274 nm (ϵ_{max}= 13.6)	λ_{max}= 331 nm (ϵ_{max}= 25)
π → π*	λ_{max}= 195 nm (ϵ_{max}= 9000)	λ_{max}= 203 nm (ϵ_{max}= 9600)

MORE conjugated double bonds MEANS LESS (*E*)is required for the electronic transition, and therefore LONGER λ at which the electronic transition occurs.

UV/Visible Spectroscopy: Instrumentation

- **Spectrometer** is an instrument which can measure the absorbance of a sample at any wavelength
- Most spectroscopic instruments in the UV/visible and IR regions are made up of five components:

1. A stable source of radiant energy
2. A wavelength selector to isolate a limited region of the spectrum for measurement
3. One or more sample containers
4. A radiation detector, to convert radiant energy to a measurable electrical signal
5. A signal-processing and readout unit consisting of electronic hardware and in modern instruments a computer.

Optical materials

- The cells, windows, lenses, mirrors, and wavelength-selecting elements in an optical spectroscopic instrument ***must*** transmit radiation in the wavelength region being investigated.

- **Sample containers:**

- Quartz or fused silica is required for the UV region (wavelengths less than 350 nm) and may be used in the visible region.

- Silicate glass is usually used for the 375–2000 nm region because of its low cost compared to quartz. Plastic cells are also used in the visible.

- The best cells have windows that are perpendicular to the direction of the beam in order to minimize reflection losses.

- The most common cell path length for studies in the UV and visible regions is 1 cm

Sample containers:

- Fingerprints, grease, or other deposits on the walls **may alter** significantly the transmission characteristics of a cell. Thus, it is important to clean cells both before and after use, and the windows must not be touched after cleaning is complete.
- Matched cells should **never be dried by heating in an oven or over a flame** because this may cause physical damage or may change the path length. Matched cells should be calibrated against each other regularly with an absorbing solution.