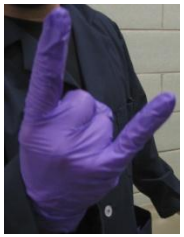
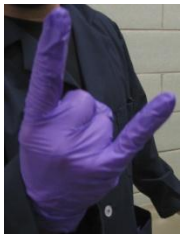
CHM 335 Experiment #1
The Absorption Spectra of Conjugated Dyes

TA: Jeff Canaria
Office: Beaupre 430A
Office Hours: Tuesday, 10:00 AM
e-mail: jcanaria@chm.uri.edu





Safety!

- The dyes used in this experiment are toxic
  - Gloves must be worn at all times when handling the dyes
  - Dilutions should be performed in the fume hood. The dyes must remain in the fume hood unless they are actively being read by the spectrophotometer.
  - Wash your hands when you are finished!

Objectives

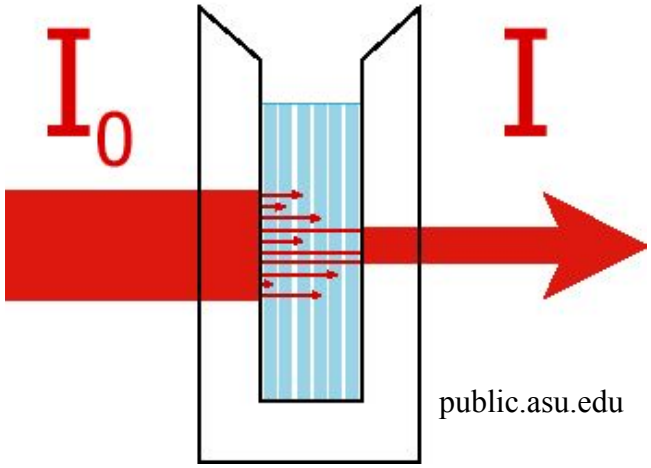
During this experiment, you will be expected to:

1. Find the maximum absorbance wavelength (λ_{max}) of each conjugated dye.
2. Calculate the theoretical maximum based on the free-electron model and compare this result to your experimental data.
3. Write a laboratory report following the specified format.

Different wavelengths are associated with different colors

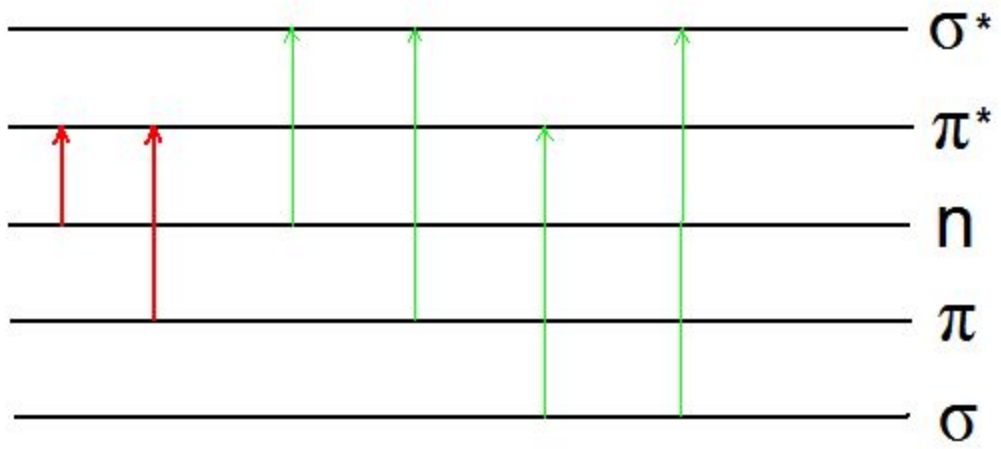
Wavelength Absorbed (nm)	Color absorbed	Color seen
400-435	Violet	Yellow-green
435-480	Blue	Yellow
480-490	Green-blue	Orange
490-500	Blue -green	Red
500-560	Green	Purple
560-580	Yellow-green	Violet
580-595	Yellow	Blue
595-605	Orange	Green-blue
605-700	Red	Blue-green

Beer Lambert law



- $A=\varepsilon cd$
- ε : molar absorption coefficient
- c: solution concentration
- d: path length
- Beer's Law only provides a good description of experimental results when c is very low!

Electronic Transitions



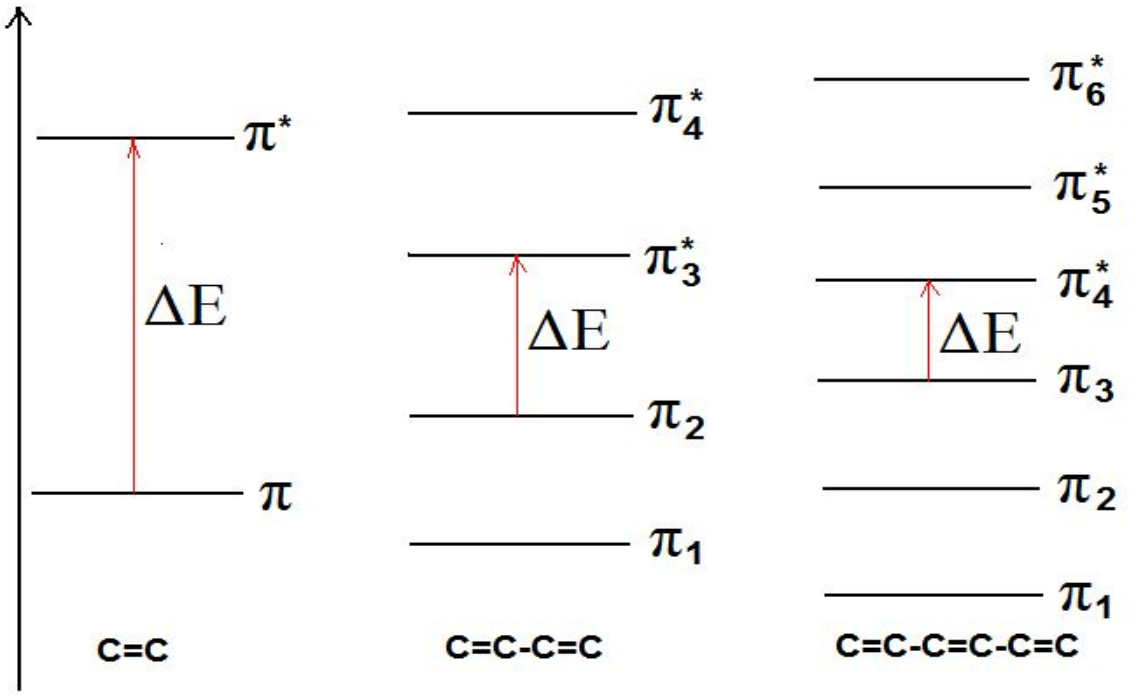
Only **n** ⟶ π* **&** π ⟶ π* can be detected by UV-Visible spectrophotometers

Conjugation (delocalization) impacts the energy required for electronic transitions

$$\Delta E = E_{photon}$$

$$E_{photon} = h \cdot c/\lambda$$

$$\Delta E = h \cdot c/\lambda$$



Derivation of the absorbance wavelength λ (nm)

1. The energy level of a particle in one-dimensional box can be written as:

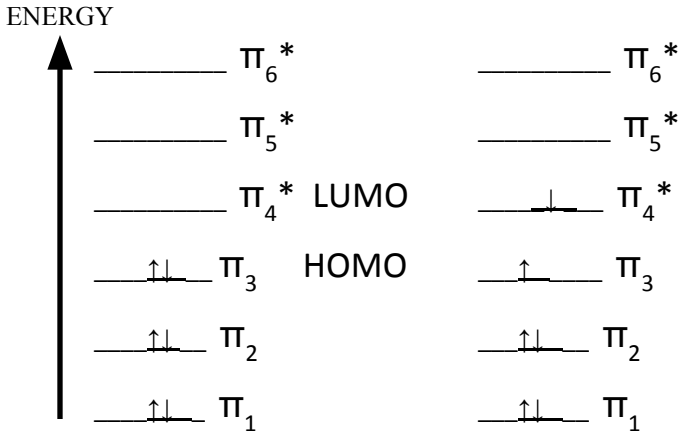
$$E_n = \frac{h^2 n^2}{8mL^2} \qquad with\ n = 1, 2, 3..$$

$m = 9.109 \times 10^{-31}\, kg =$ mass of electron,

$h = 6.626 \times 10^{-34}\, J\ s =$ Plank's constant

$n =$ quantum number, L : Length of box

2. Absorption of light by a molecule or ion is associated with a one – electron jump from the HOMO (n_1) to the LUMO (n_2). Where N is an even number of π electrons, $\mathbf{n_1 = N/2}$, and $\mathbf{n_2 = (N/2)+1}$.



3. The energy change for the transition from HOMO to LUMO can be written as:

$$\Delta E = E_{n_2} - E_{n_1} = \frac{h^2 n_2^2}{8mL^2} - \frac{h^2 n_1^2}{8mL^2} \xrightarrow[n_1=\frac{N}{2}]{n_2=\frac{N}{2}+1} \frac{(N+1)}{L^2}\left(\frac{h^2}{8m}\right)$$

4. Since $\Delta E = E_{photon} = h\upsilon = hc/\lambda$, where : λ= wavelength,
c = the speed of light ($2.99792458*10^8 ms^{-1}$)

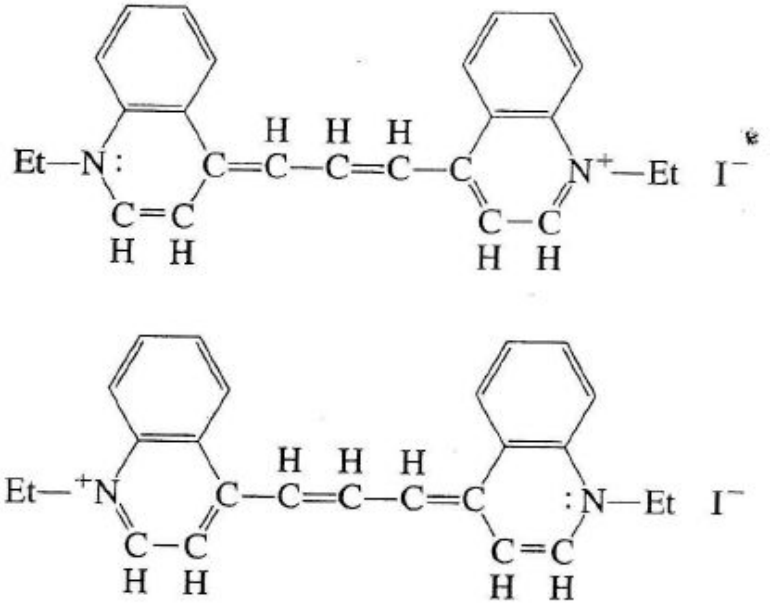
$$\Delta E = \frac{hc}{\lambda} = \frac{(N+1)}{L^2}\left(\frac{h^2}{8m}\right)$$

$$\lambda = \frac{L^2}{N+1}\left(\frac{8mc}{h}\right)$$

5. Let's let p equal the number of carbons in the polymethine chain, then **N = p + 3.**
(3 = the π electrons from the nitrogen atoms).

6. Using the approximation that L is the length of the chain between nitrogen atoms plus one bond distance on each side gives us L = (p + 3)l, where l is the bond length between atoms. The equation can now be written as

$$\lambda = \frac{L^2}{N+1}\left(\frac{8mc}{h}\right) \xrightarrow[N=p+3]{L=(p+3)\ell} \frac{(p+3)^2}{p+4}\left(\frac{8mc\ell^2}{h}\right)$$



7. Substituting 0.139 nm (the bond length in benzene) in for ℓ, plus the values for h,c,m, & N gives us

$$\lambda(\text{in } nm) = \left(\frac{8mc\ell^2}{h}\right)\frac{(p+3)^2}{p+4} = (63.7)\frac{(p+3)^2}{p+4}$$

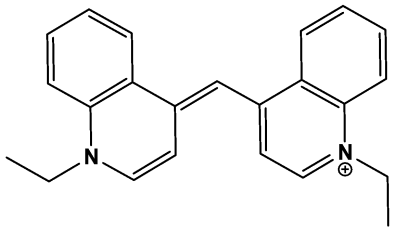
8. The box length L can be increased when there are easily polarizable groups at the ends of a carbon chain. Taking this into account gives the following formula

$$\lambda(\text{in } nm) = (63.7)\frac{(p+3+\alpha)^2}{p+4}$$

For the dye we will use as a standard, α should lie between 0 & 1.

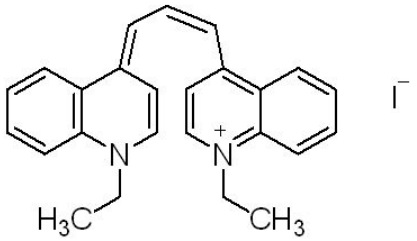
Experimental

Three dyes will be used in this experiment:



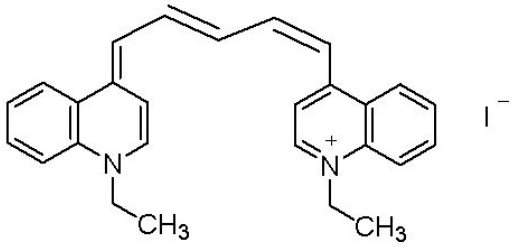
1,1'-diethyl-4,4'-cyanine iodide

Dark Blue



1,1′-Diethyl-4,4′-carbocyanine iodide

Blue



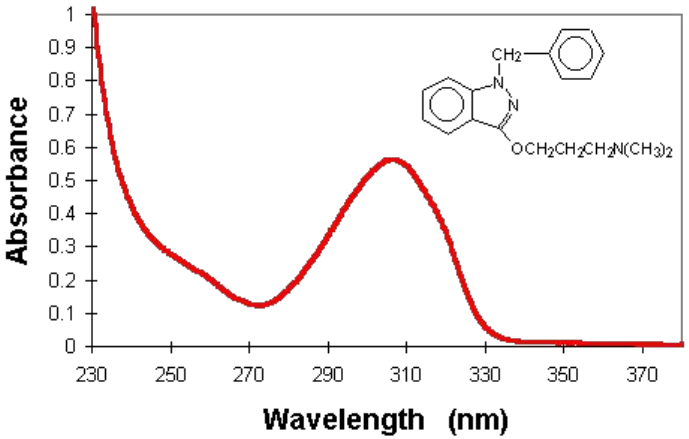
1,1′-Diethyl-4,4′-dicarbocyanine iodide

Green

Each dye is in methanol solution of with a concentration of approximately 1 x 10^{-4} M.

Procedure

1. Fill a cuvette with about 2 mL of sample (dye) solution. The dyes can be run in any order.
2. Use the spectrophotometer to find the absorbance maximum of the dye
3. If necessary (i.e. the graph does not show a smooth peak and/or the maximum absorbance number exceeds 1.0), dilute the sample by removing **half the volume of the dye** and replacing it with an equal amount of methanol. Reread the maximum absorbance. Repeat the dilution process until the maximum absorbance is below 1. Record both λ_{max} **and the absorbance values of all the samples**. Select one that has Absorbance between 0.5 to 0.8 to read off λ_{max}.
4. Repeat the above process with the other two dyes. Remember to record both λ_{max} **and the number of dilutions**.



Moore, D.E.; Wang, J.
www.photobiology.com/v1/moore/index.htm

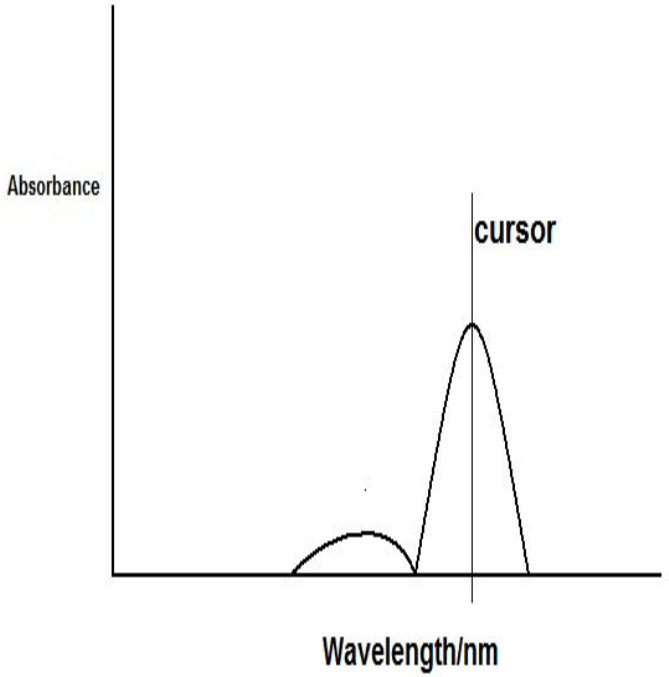
Procedure

1. Pipet about 2ml of pure methanol into a cuvette (approximately 2/3 to ¾ of the cuvette).
2. Insert the reference cuvette (methanol) into the rear cuvette holder. Do not insert the cuvette for the dye yet.
3. Click the "Baseline" button in the bottom left toolbar of the UV-Probe 2.51 software.
4. Allow the scan to finish (note that the wavelength being scanned changes during this process).
5. Pipet about 2ml of one of the dyes into a second cuvette.
6. Insert the cuvette containing the dye into the front cuvette holder.
7. In the spectrum window, make sure that the "Active" tab is selected and then click the "Start" button in the bottom left toolbar of the software.
8. Allow the scan to finish.
9. Right-click in the spectrum window and hover the mouse over the "crosshair" option and then move the mouse cursor over to "display."
10. Hover the cursor over the maximum absorbance peak and record the corresponding wavelength (x-axis) and absorbance (y-axis).
11. If the absorbance reading is not between 0.5 and 0.8 then dilute the sample as mention previously until an absorbance reading that falls within acceptable range is obtained.
12. Repeat steps 5-11 for the remaining dyes.

A Note on Dimerization

The dye solutions may dimerize, which can lead to a shoulder on the absorption peak. The extent of dimerization will vary depending upon the solution concentration.

You should use the larger peak in the spectrum; this absorption should be due to the monomeric form that you want to study.



Calculations

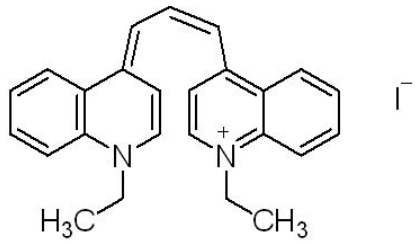
You will be comparing your experimental results to the theoretical values obtained from the free-electron model calculation discussed earlier:

$$\lambda(\text{in } nm) = (63.7)\frac{(p+3+\alpha)^2}{p+4}$$

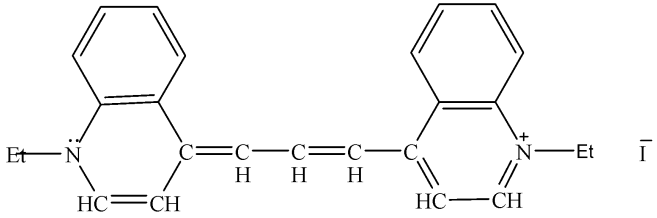
1. Calculate α by plugging in the λ max value that you measured for **1,1'-Diethyl-4,4'-carbocyanine iodide. [Blue Dye!]**
2. Using your calculated α value, determine λ max for the remaining dyes.
3. Compare the calculated λ max values with the values that you obtained experimentally.

Sample Calculation

1. If we use 1,1'-Diethyl-4,4'-carbocyanine iodide as an example, then p = 9.



1,1'-Diethyl-4,4'-carbocyanine iodide



2. If $\alpha = 0.01$*, solving the equation

$$\lambda \text{ (in nm)} = 63.7\ ((p + 3 + \alpha)^2/(p + 4))$$

gives us

$$\lambda = 63.7((9 + 3 + 0.01)^2/(9 + 4)) = 705.8 \text{ nm}$$

* **This number was chosen for illustrative purposes ONLY. It is NOT the value that you will use for α. Your value for α must be based on 1,1'-Diethyl-4,4'-carbocyanine iodide.**

Laboratory Reports

- An outline of what is expected in your report for the first laboratory experiment can be found in your course syllabus. (**p. 14-15**) In addition to what is described in the syllabus:
  - Include the number of dilutions for each dye in your data table.
  - In the Summary & Discussion section, use Beer's Law to explain why the samples needed to be diluted. Based on the number of dilutions required for each dye, answer the following question: If you were using a HPLC/UV instrument to study these dyes, which dye would have the lowest limit of detection?
- You are responsible for reading this outline and writing a report that contains all of the required components.
- Your original data must be included with your report.

Laboratory Reports Con't

- The 7 general sections of your laboratory report are:

1. Cover page
2. Introduction
3. Theory
4. Signed Procedure
4. Signed Original Data
5. Calculations
6. Summary & Discussion

- Make sure that your cover page clearly indicates the author of the lab report.
- Points WILL BE DEDUCTED for Poor English, including poor grammar and spelling errors!!!
- *Reports will be collected in lab next week*. Remember the lateness policy!!

Sample Cover Page

Experiment 1: The Absorption Spectra of Conjugated Dyes

September 12, 2018

My Name Is: Jeff Canaria

My Lab Partner's Name Is: Sze Yang