PHOTOPHYSICS & MOLECULAR SPECTROSCOPY
By Prof. Ayben Kilislioglu and Sanja Tepavcevic, adapted from A.M. Halpern,
Experimental Physical Chemistry: A Laboratory Textbook, 2nd Ed.

Experiment 34
Excited State Properties of 2-Naphthol

1. Introduction

Molecular fluorescence spectrometry is a very powerful method for analysis of organic compounds because high sensitivity and high selectivity can frequently be achieved [1]. A spectrofluorimeter (Figure 1) is the instrument of choice for recording fluorescence spectra. A beam of monochromatic light excites the specimen in the cell, and the emission is observed and analyzed at right angles to the incident beam. Fluorescence spectral data are generally presented as emission spectra, which are plotted as the fluorescence intensity versus wavelength or wave numbers [2,3].

The electronic structure of a molecule determines such physical and chemical properties as its charge distribution, geometry (therefore dipole moment), ionization potential, electron affinity, and, of course, chemical reactivity. If the electronic structure of a molecule is changed, therefore, we would expect its physical and chemical properties to be altered. Such a rearrangement in electronic structure can, in fact, be brought about (and very rapidly, \(10^{-13}\) s) if the molecule is raised to an electronically excited state via the absorption of a quantum of light (photon) whose energy matches the gap between the molecular ground and excited state energy levels[4].

In this experiment, you will study the fluorescence of aqueous solutions of 2-naphthol (ArOH) as a function of pH and determine some ground and excited state properties of this organic molecule.

![Figure 1. Schematic diagram of a spectrofluorimeter. Note that the Ocean Optics spectrofluorimeter does not have an excitation monochromator and instead uses broadband radiation to excite the sample. (Figure from J.A. Barltrop and J.D. Coyle, Excited States in Organic Chemistry, Wiley:1975, p.71).](image-url)
II. Theory

Some molecules will fluoresce or emit light after you excite them from their ground state to an excited electronic state. The most common method of inducing this excitation is optically, by absorption of light. Fluorescence can occur when the molecule relaxes back down to its ground state, but this is not the only process that can occur for an excited molecule. Furthermore, not all light emission from excited molecules is due to fluorescence. These details will be clarified below. This lab report concerns itself with fluorescence and optical absorption events that occur in the ultraviolet and visible (UV/Vis) regions of the spectrum.

Characteristics of fluorescence emission

1) Stokes' shift

Except for atoms in the vapor phase, one invariably observes a shift to lower wavelength (i.e., a loss of energy) of the emission relative to the absorption. Energy losses between excitation and emission are observed universally for fluorescing molecules in solution. An unshifted emission is observed when the gas concentrations are sufficiently small so that the excited molecule does not collide with any other molecules prior to emission.

2) Invariance of the emission spectrum with excitation wavelength

The relaxation occurs in about $10^{-12}$ s, and is presumably a result of a strong overlap among numerous states of nearly equal energy. Because of this rapid relaxation, emission spectra are usually independent of the excitation wavelength.

3) Mirror image rule

Generally, the fluorescence emission spectrum appears to be a mirror image of the absorption spectrum, here the absorption representing the ground state singlet $S_0$ to excited state triplet $S_1$ transition. An electronic state of a molecule can be characterized by the total spin magnetic quantum number $M_S$ and total spin angular momentum quantum number $S$. If $m_s$ is the spin quantum number of an individual electron with values of either $+\frac{1}{2}$ or $-\frac{1}{2}$ then $M_S = \sum(m_s)$, the sum of the values of $m_s$ of the individual electrons. Just as there are $2l + 1$ values of $m_l$ for the orbital angular momentum of the electron in a hydrogen atom there are $2S + 1$ values of $M_S$ for a molecule:

<table>
<thead>
<tr>
<th>$S$</th>
<th>$M_S$</th>
<th>$2S + 1$</th>
<th>multiplicity</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>1</td>
<td>singlet</td>
</tr>
<tr>
<td>1/2</td>
<td>+1/2 or -1/2</td>
<td>2</td>
<td>doublet</td>
</tr>
<tr>
<td>1</td>
<td>-1, 0, or 1</td>
<td>3</td>
<td>triplet</td>
</tr>
<tr>
<td>3/2</td>
<td>-3/2, -1/2, 1/2, or 3/2</td>
<td>4</td>
<td>quartet</td>
</tr>
<tr>
<td>2</td>
<td>-2, -1, 0, 1, or 2</td>
<td>5</td>
<td>quintet</td>
</tr>
</tbody>
</table>

The symmetric nature of this spectrum is a result of the same transitions being involved in both absorption and emission, and the similarities among the vibrational energy levels of $S_0$ and $S_1$. In many molecules these energy levels are not significantly altered by the different electronic distributions of $S_0$ and $S_1$. According to the Franck-Condon principle all electronic transitions are vertical, they occur without change in position of the nuclei. This is because electronic transitions are much faster than vibrational or rotational transitions and occur before the molecule can change its geometry. As a result, if a particular transition probability (Franck-Condon Factor) between vibrational levels is largest in absorption, the reciprocal transition is also most probable in emission (Figure 2)[3].
An Ideal Spectrofluorimeter

We wish to record excitation and emission spectra that represent the relative photon intensity per wavelength interval. To obtain such “corrected” emission spectra the individual components must have the following characteristics [3]:
1. The light source must yield a constant photon output at all wavelengths.
2. The monochromator must equally transmit all wavelengths in the emission spectrum.
3. The monochromator efficiency must be independent of polarization.
4. The detector (photomultiplier tube or CCD array or Si diode) must detect photons of all wavelengths with equal efficiency.

Unfortunately, light sources, monochromators, and detectors with such ideal characteristics are not available. As a result, one is forced to compromise on the selection of components and to correct for the non-ideal response of the instrument.

An absorption spectrophotometer contains these same components, and one may wonder why it is possible to record correct absorption spectra [3].

Processes of Deactivation

Electronic excited molecules can relax by emitting light (i.e., fluorescence) in a process which is generally referred to as a radiative transition. Relaxation can also occur nonradiatively, that is by relaxation processes that do not emit light.

Molecules in excited vibrational states rapidly dissipate their excess vibrational energy and relax to the ground vibrational level in a given electronic state. The energy goes into thermal or vibrational motion of the solvent molecules in condensed phases. This nonradiative process is denoted vibrational relaxation. This typically takes $10^{-11}$ to $10^{-10}$s. Since a typical vibrational period is $10^{-13}$s, many vibrations occur before the excess vibrational energy is lost.

For most organic molecules that contain an even number of electrons, the ground state is characterized by having all electron spins paired; the net spin angular momentum is zero and such an arrangement is called a singlet state. When considered in terms of molecular orbitals (MO), electronic excitation involves the promotion of an electron from a filled MO to a higher, vacant MO. This new orbital configuration, which characterizes the electronically excited state, may be one in which the two electrons in the singly occupied MOs have opposite spins. Accordingly, this electronically excited state is also a singlet.
The ground, and lowest electronically excited, singlet states are often denoted as $S_0$ and $S_1$, respectively. Higher excited singlet states are referred to as $S_2$, $S_3$, ......,$S_n$. This experiment deals with excited singlet states [4].

The crossover between two states of the same multiplicity (same spin quantum number or paired/opposite spins such as singlet to singlet) is a nonradiative electron state transition called internal conversion. Fluorescence is a radiative transition between electronic states of the same multiplicity [6]. Generally, direct transitions between singlet and triplet states are not allowed (this is often called spin forbidden) or only occur much less frequently than spin allowed transitions (i.e., singlet to singlet).

For most molecules, the electrons are paired in the ground state so that fluorescence involves a singlet-singlet transition. Because internal conversion to $S_1$ and vibrational relaxation are more rapid processes than fluorescence, fluorescence usually occurs from the ground vibrational state of $S_1$ to various vibrational levels in $S_0$ ($S_1 \rightarrow S_0 + \text{hv}$). For this reason, only one fluorescence band is normally observed even if absorption to different excited sites occurs. Typically, fluorescence requires $10^{-10}$ to $10^{-6}$ s to occur. Fluorescence usually appears at longer wavelengths than absorption because absorption transitions are to higher excited electronic states or to higher vibrational levels in the $S_1$ manifold. The term external conversion refers to nonradiative processes in which excited states transfer their excess energy to other species, such as solvent or solute molecules [6].

Part of the energy level diagram for a hypothetical aromatic molecule is shown below in a Jablonski diagram (Figure 3) [6].

![Figure 4. Deactivation processes for an excited molecule: (a) absorption; (b) vibrational relaxation; (c) internal conversion; (d) fluorescence; (e) external conversion; (f) intersystem crossing; (g) phosphorescence. (Figure from J.D. Ingle, Jr and S.R. Crouch, Spectrochemical Analysis, Prentice-Hall:1988, p.338).](image-url)
Absorption transitions to triplet states from ground states which are singlet are forbidden by symmetry, although weak absorption is possible in some molecules. The triplet state can also be populated from excited singlet states by intersystem crossing, which is a crossover between electronic states similar to internal conversion except that the states have different multiplicities (usually $S_1 \rightarrow T_1$) [6].

After intersystem crossing, a molecule in the $T_1$ state deactivates by vibrational relaxation to the ground vibrational level of $T_1$. Normally, the triplet state deactivates by external conversion or intersystem crossing to the ground state ($T_1 \rightarrow S_0$). The triplet state can also deactivate by emission of a photon. This (radiative) deactivation process between electronic states of different multiplicity is called phosphorescence (typically $T_1 \rightarrow S_0 + \text{hv}$). Usually, phosphorescence is very slow, taking anywhere from $10^{-4}$ to $10^4$ s to occur because the process is spin forbidden [6].

If the energy of the excitation photon is greater than the convergence limit of the excited electronic state, a bond is ruptured after absorption. This process is called dissociation. Dissociation is more likely in molecules that absorb at wavelengths shorter than 200 nm (200 nm corresponds to 586 kJ/mol) [6].

Although measurements of the physical and chemical properties of a molecule in its ground state can be carried out, more or less, at leisure (assuming that the molecule is thermally stable), the examination of these properties in its excited states is severely hampered by the fact that these states are very short lived. For most molecules, $S_1$ states have lifetimes ranging from $10^{-6}$ to $10^{-11}$ s. Excited states are metastable; they undergo decay processes that dissipate the energy they possess relative to more stable products. For example, the excited state of a molecule may, in general; spontaneously return to the ground state via photon emission (fluorescence); convert electronic excitation into ground state vibrational energy (heat); or undergo bond dissociation or rearrangement, possibly leading to a change in electron spin multiplicity. Because spontaneous emission from an excited state (i.e., fluorescence) often takes place very rapidly, fluorescence can be used as a probe, or measurement, of excited state concentration (e.g., fluorescence assay). In addition, fluorescence studies can provide information about the physical and chemical properties of these short-lived singlet states. This field of experimentation is called photophysics [4].

![2-naphthol](image)

2-naphthol (ArOH)

In aqueous solution, ArOH behaves as a weak Bronsted/Lowry acid, forming the hydronium ion and its conjugate base, the naphthoxy ion, $\text{ArO}^-$. 
\[
\text{ArOH} + H_2O \leftrightarrow \text{ArO}^- + H_3O^+
\]

It is instructive to measure the acidity constant of ArOH in its lowest excited electronic state, denoted as $K_a^*$, and to compare this value with that of the ground state, $K_a$. This
information indicates how the change in electronic structure alters the charge density at the oxygen atom.

**Calculations of \( K_a \)**

Because the bulk 2-naphthol concentrations \( [ArOH]_0 \) are identical in each of the solutions studied, the following material balance applies:

\[
[ArOH]_0 = [ArOH] + [ArO^-].
\]  

(1) 

Under the condition that both the free acid and conjugate base absorb at \( \lambda_{\text{max}} ArOH \), the wavelength of maximum absorption for \( ArOH \), and that Beer’s law holds:

\[
A(\lambda_{\text{max}} ArOH) = \varepsilon_{ArOH} [ArOH] + \varepsilon_{ArO^-} [ArO^-].
\]  

(2) 

where \( A \) is the absorbance (for a 1-cm path length), and \( \varepsilon_{ArOH} \) and \( \varepsilon_{ArO^-} \) are the molar absorptivity coefficients of the free acid and conjugate base at \( \lambda_{\text{max}} (ArOH) \), respectively.

From the lowest pH solution obtain value for the \( \varepsilon_{ArOH} \) and from the highest pH solution value for \( \varepsilon_{ArO^-} \) and use these values for calculations at all other pH values (see as an example Figure 6 showing fluorescence spectrum of different pH).

![Fluorescence spectrum of 2-naphthol in solutions of different pH](image)

Figure 3.11 Fluorescence spectrum of 2-naphthol in solutions of different pH (from ref. 34). (1) 0.02 M NaOH, (2) 0.02 M sodium acetate + 0.02 M acetic acid, (3) pH 5-6, (4) 0.004 M HClO₄, (5) 0.15 M HClO₄

**Figure 6.** Fluorescence spectrum of 2-naphtol in solutions of different pH
Furthermore, combining equations (1) and (2) produces
\[
A(\lambda_{\text{max}} \text{ArOH}) = \left[ \epsilon_{\text{ArOH}} - \epsilon_{\text{ArO}^-} \right] [\text{ArOH}] + \left[ \epsilon_{\text{ArO}^-} [\text{ArOH}] \right]_0
\]  
(3)

This relation allows \([\text{ArOH}]\) to be determined under equilibrium conditions. Once this is known, the value of \([\text{ArO}^-]\) at the same pH can be obtained from equation (1). Because the acidity constant is
\[
K_a = \frac{[H_3O^+] \text{ArO}^-}{[\text{ArOH}]},
\]  
(4)

(assuming that the activity coefficient ratio is unity in these dilute solutions), you can obtain the \(pK_a\) value for ArOH from a graphical analysis of
\[
pH = pK_a + \log \left( \frac{[\text{ArO}^-]}{[\text{ArOH}]} \right),
\]  
(5)

\[
pK_a = pH - \log \left( \frac{[\text{ArO}^-]}{[\text{ArOH}]} \right)
\]  
(6)

using linear least squares regression. Report the least-squares value of \(pK_a\) along with its standard deviation. Compare your results with the literature values.

**Calculations of \(K_a\)**

The experimental method is best introduced in terms of the energy-level diagram shown in Figure 5. The relative energies of the free acid and its conjugate base (the naphthoxy ion) are indicated for both the electronic ground (\(S_0\)) and lowest excited (\(S_1\)) states in aqueous solution. Each anion is elevated with respect to its free acid by an energy, \(\Delta H\) and \(\Delta H^*\), respectively. These are the enthalpies of deprotonation. Both the ground state acid and its conjugate base can be transformed to their respective excited states via the absorption of photons of energy \(h\nu_{\text{ArOH}}\) and \(h\nu_{\text{ArO}^-}\). For simplicity, these absorption transitions are shown to be equal to the fluorescence from the excited to the ground states of the acid and conjugate base[4]. (The ground and excited state vibrational levels involved in the transitions are not indicated.)
We can express the free energy of deprotonation of ArOH in terms of the enthalpy and entropy of deprotonation and the equilibrium (ionization) constants:

\[ \Delta G^o = \Delta H^o - T \Delta S^o = -RT \ln K_a, \]  

(7)

and

\[ \Delta G^* = \Delta H^* - T \Delta S^* = -RT \ln K_a^*, \]  

(8)

for the \( S_0 \) and \( S_1 \) states, respectively. If we make the assumption that the entropies of dissociation of ArOH and (ArOH)* are equal, it follows that

\[ \Delta H^* - \Delta H = -RT \ln \left( \frac{K_a^*}{K_a} \right), \]  

(9)

and thus from Figure 5, it can be deduced that

\[ \Delta H + N_A h \bar{\nu}_{ArO^-} - \Delta H^* = N_A h \bar{\nu}_{ArOH}, \]  

(10)

where \( h \) is Planck’s constant. Avogadro’s number, \( N_A \), has been included to put each energy term on a molar basis. Combining equations (9) and (10) and rearranging gives

\[ - \ln \left( \frac{K_a^*}{K_a} \right) = \frac{N_A h \left( \bar{\nu}_{ArO^-} - \bar{\nu}_{ArOH} \right)}{RT}. \]  

(11)

Thus knowledge of the energy gap between the ground and first excited states for both the free acid and its conjugate base leads to an estimate of \( K_a^* \), if \( K_a \) is known. The analysis presented above accounts for the observed thermodynamic and spectroscopic energy differences. This approach is often referred to as a Förster cycle, named after the scientist who invented it in 1949. Equation (11) can be recast into a more convenient form.

\[ pK_a^* = pK_a - \frac{N_A hc}{2.303RT} \left[ \bar{\nu}_{ArOH} - \bar{\nu}_{ArO^-} \right], \]  

(12)
Where \( c \), the speed of light, has been incorporated in the expression to convert the transition energies of ArOH and ArO' into wavenumbers (\( \tilde{\nu} \) in units of cm\(^{-1}\)), a common spectroscopic energy unit. The acidity constants are expressed as pK values. The 2.303 is needed in the denominator because the pK's are defined as \(-\log_{10}K\).

The question now is how to obtain the spectroscopic energy difference (\( \tilde{\nu}_{ArO'} - \tilde{\nu}_{ArOH} \)), or \( \Delta \tilde{\nu} \), pertinent to the Förster cycle in 2-naphthol. Three approaches can be considered. The first is to base the measurement on the absorption maxima of the free acid and conjugate base. The second is to use the fluorescence maxima of the two species, and the third is to use what is called the 0-0 energies of ArOH and ArO'. The first two methods are somewhat more straightforward. Obtaining \( \Delta \tilde{\nu} \) from absorption data has the advantage that the energy difference that is obtained does not require instrumental correction. Although obtaining \( \Delta \tilde{\nu} \) from the fluorescence maxima seems simple enough, many fluorimeters produce fluorescence spectra that are distorted by the wavelength response of the monochromator/photomultiplier combination. Thus unless these spectra are corrected for this sensitivity distortion, the recorded fluorescence maxima will depend (although perhaps subtly) on the particular instrument used and thus will not represent the "true" or absolute spectroscopic properties of the ArOH/ArO' system.

The third approach provides the value of \( \Delta \tilde{\nu} \) that best represents the energy difference between \( S_1 \) and \( S_0 \) implied in the Förster cycle, \( \Delta \tilde{\nu}_{0-0} \). Unfortunately \( \Delta \tilde{\nu}_{0-0} \) cannot be determined directly in all cases (such as ArOH), but it can be estimated from an analysis of both the absorption and fluorescence spectra. This is shown schematically in Figure 6 for a single species (ArOH or ArO').

The absorption and (preferably instrument-corrected) fluorescence spectra are both plotted on a common energy axis. Furthermore, the spectra are presented so that they are normalized to have identical maxima. The point of intersection of these spectra can be approximated as the 0-0 energy gap between \( S_0 \) and \( S_1 \). This approach does not apply to those cases in which the \( S_0\text{-}S_1 \) transition is distorted by a nearby (and especially more intensely absorbing) \( S_0 \) to \( S_1 \) transition.

Whatever method is used to determine \( \Delta \tilde{\nu} \), it should be used consistently with both species, ArOH and ArO'.

**Figure 6.** Schematic diagram of the absorption and fluorescence spectra of a molecule. It is assumed that these transitions are between the same two electronic states, i.e., \( S_0 \leftrightarrow S_1 \). (Figure from A.M. Halpern, Experimental Physical Chemistry, A Laboratory Textbook, 2nd Ed.).
Instrumentation

For fluorescence spectroscopy, the energy source must be in the ultraviolet range. Hydrogen, deuterium, or mercury lamps may be used, but xenon arcs are perhaps the most popular. Quartz optics and cells are required, and since the fluorescent radiation is in the visible region, Si detectors commonly used on modern instruments. Although these components are all part of an ultraviolet spectrophotometer, there are differences between this instrument and a fluorimeter [5].

Fluorescent radiation is emitted in all directions, and advantage is taken of this fact in designing the fluorimeter. It is possible to measure the amount of fluorescence that comes from the sample in the same direction as the incident radiation, but this measurement would require a very good monochromator and is difficult to do with accuracy. By moving the detector at right angles to the cell, only the fluorescent radiation is measured, and much more accurate results can be obtained [5].

Factors influencing results

1. Since some compounds are destroyed by the intense ultraviolet radiation that is used to excite the molecules, the slow analyst may find it difficult to obtain reproducible results.

2. The adjustment of pH is very important. Since the protons can protonate the nonbonding electrons, thereby making them no longer nonbonding, the fluorescence wavelengths can be shifted considerably and may even be destroyed. It is essential to take great care to adjust the pH properly when making determinations involving fluorescence.

3. Temperature is another factor which may affect results. One of the difficulties with fluorescing molecules is that they don’t all behave as we want them to, and most of them lose their energy by collision, rather than by fluorescing. If we could stop the collisions or at least reduce them, then the fluorescence intensity would increase. A good method to reduce molecular collision is to solidify the material by freezing it. (If the sample and cell are placed in dry ice/acetone, or better yet, liquid nitrogen, the sample is frozen in a few seconds, and in many instances the fluorescence intensity increases by as much as 10 to 1000 times. Readings must be made in a few seconds after the cell is removed from the freezing solution, since the cold cell will freeze moisture from the air onto its surface and ruin the analysis.)

4. Self-absorption is a problem at high sample concentrations. The emitted fluorescent radiation from one molecule is also just the right energy to be absorbed by a second molecule. This radiation may then be lost by further collision, or it may be reradiated as radiation. If it is reradiated, it has gone through two molecules, yet the detector would count it as having come from one molecule, and low results would be obtained. This is a serious problem. Only dilute solutions will alleviate this effect and follow Beer’s law [5].
III. Experimental protocol

Safety precautions
- Always wear safety goggles in the laboratory. Ultraviolet light-absorbing eye protection is required. Some plastic safety goggles do not completely absorb ultraviolet radiation.
- 2-Naphthol is an irritant. If you prepare solutions from solid material, you must wear gloves; if possible, work in a fumehood.
- Be sure you have been instructed to use the proper pipetting techniques when handling 2-naphthol solutions. Never pipet by mouth.
- If you are to obtain fluorescence spectra, be sure that any ozone produced by the ultraviolet source is vented. Ozone is a noxious, dangerous gas that has an acrid odor. If you detect this gas, leave the vicinity, inform your instructor, and increase air circulation at once.
- Waste solvents should be disposed properly, as directed by your TA.

Step I: Absorption

1) Obtain absorption spectra of all five solutions of ArOH (2 × 10^{-4} M; the actual concentration must be accurately known). In one solution, the free acid must predominate (low pH), and in another, the conjugate base must be the major naphthol component (high pH). Use concentrated stock solutions of HCl and NaOH e.g., 0.10 M) to create [H^+] and [OH^-] of 0.02 M, respectively. It is important that the ArOH concentration be the same in each case. Label and save these solutions. Record these spectra on the same chart paper having a common wavelength axis so that the spectra overlap. You also need to record the spectra of all the buffer solutions.

2) Now, obtain absorption spectra of ArOH (also 2×10^{-4} M) at intermediate pH values. Adjust pH levels by using ammonium chloride buffer solutions (NH₄OH/NH₄Cl), for example, 0.1 M/0.1 M, 0.1/0.2, or 0.2/0.1. These solutions can be conveniently prepared from 1.00 M stock solutions of NH₄OH and NH₄Cl. Obtain at least three spectra, run over the entire wavelength range as in step 1, illustrating both free acid and conjugate base absorption. If necessary, choose appropriate ratios of the NH₄OH and NH₄Cl stock solutions to produce a satisfactory series of ArOH/ArO^- spectra. It is essential that the 2-naphthol concentrations be identical (and accurately known) in each case. Immediately after recording each spectrum, measure the actual pH of the solution using a properly calibrated pH meter. Label and save these solutions.

3) It is instructive to overlap each spectrum on the same strip-chart paper, making sure that the wavelength is synchronized in all cases. Make sure that the temperature of the samples is constant (to within 1°C) or otherwise controlled throughout the experiment. If the bulk ArOH concentration, e.g., [ArOH]+[ArO^-], is invariant, the spectra, when properly overlapped, should intersect at a common wavelength called the isobestic point (equal absorption). The presence of an isobestic point indicates that there is a closed system (as a function of the variable pH) consisting of two species in equilibrium.
UV/VIS Operation

OCEAN OPTICS RED TIDE UV/VIS OPERATING INSTRUCTIONS

• to use system

TURN ON POWER STRIP; IMPORTANT -
UV-VIS has power before computer
has booted to initialize properly
TURN ON COMPUTER

• Log on - student, password: pchem (lower case)
• Launch LOGGER PRO 3.6.1
• Goto Experiment > Calibrate > spectrometer 1 (follow instructions)
• Goto Experiment Start DATA Collection
• Goto Experiment Stop DATA Collection
• Save DATA or Export DATA as (Text or Inspire Data CVS)
• Exit Logger Pro
• to shut system down

TURN OFF COMPUTER
TURN OFF POWER STRIP; IMPORTANT -
this removes power from UV-VIS

Step II: Fluorescence

3) Using the same solutions from Step 1, obtain the fluorescence spectra of naphthol and its conjugate base. If possible, correct these for the monochromator/photomultiplier response of the fluorimeter.

Fluorescence Operation, Ocean Optics/ Saving data in ASCII file
• Start the program Ocean Optics.
• Use the dark light bulb icon. Then select File/Save/Dark to save permanently.
• Place the sample in the cell holder.
• Turn on the light source. (on/off switch)
  o NOTE: do not touch other switch. Flash mode should be on Multiple.
• Check the box strobe enable leave the box at 100 ms.
• The system is now Acquiring Data. Adjust integration time for full scale.
  o (NOTE: you should keep same integration time for all pH)
• Adjust X-axis. **View/Spectrum scale/Set scale.** Type in value to center spectra on X-axis.

• Subtract the dark spectrum from the data. Use the **subtract dark icon**.

• Save data under **File/Save/Processed**.
  (NOTE: This will save the file in the form that you can edit the file using **Wordpad**)

• To overlay samples go to **overlay/select add overlay then file**.

• Select **File/save/experiment** to save complete set of acquisition and processing parameters for the active spectral window.

• **File/print** or use the **print icon** (HP laserjet 1022n)

• Uncheck the **strobe enable** and move the light source power switch to the **right** when finished.

**Step III: Plotting of Data**
You should download the data on a diskette so you can export the data from two different instruments and plot onto the same graph using Excel (or some other plotting program).

**Step IV: Determination of K_a and K_a**
From the high and low absorption spectra, determine \( \lambda_{\text{max}} \) ArOH and the molar absorptivity coefficients of ArOH and ArO\(^-\) at this wavelength. Using these values, along with the absorbance (pH) data, determine pK_a for ArOH using equations (1), (3) and (6).

Calculate pK_a\(^\star\) for (ArOH)\(^\star\) from the pK_a value obtained and from whatever method(s) you can use to determine \( \left( \bar{v}_{\text{ArO}} - \bar{v}_{\text{ArOH}} \right) \). Compare these values and perform an error analysis. Discuss the errors that the primary measurements have on the derived value of pK_a\(^\star\).

Other molecules that can readily be studied using this procedure are 2-naphthoic acid, acridine, and quinoline.

![2-Naphthoic acid](image)

![Acridine](image)

![Quinoline](image)
Questions and Further Thoughts

1. In comparing the values of $K_a$ and $K_a^*$, what can you deduce about the change in electron density at the O atom in 2-naphthol in the electronically excited state relative to the ground state?

2. If $pK_a^* < pK_a$ (i.e., the excited state species is a stronger acid), can you comment on the relative (absolute) magnitudes of the enthalpies of deprotonation? See Figure 4.

3. On what basis can we justify the assumption that the entropies of deprotonation of the ground and excited state of 2-naphthol molecules are equal [see equations (1)-(3)]? Can you think of another possibility in which this assumption is a poor one?

4. Other molecules that can be studied using this technique are 2-naphthoic acid, acridine, and quinoline (see above). Indicate the protolytic reactions for these molecules, i.e., write the aqueous acid-base reactions.

5. Can you predict before doing an experiment whether the excited state of a molecule is a stronger or weaker acid relative to its respective ground state? What information would you need to perform such an assessment?

6. Give for both the acid and base forms: $\lambda_{\text{max}}$ for absorption, $\lambda_{\text{max}}$ for fluorescence, and the 0-0 energy gap that you observed.

7. Clearly define what an isobestic point is and give your experimental value.

8. Do your fluorescence spectra have a point comparable to the isobestic point? If so, what is the experimental value?

References


4) A.M. Halpern, Experimental Physical Chemistry, A Laboratory Textbook, 2nd Ed..
