

Reaction Kinetics by Time-Resolved Laser Spectroscopy

Physical Chemistry Laboratory: Chem. 329

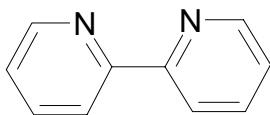
Spring 2002

Introduction:

In this experiment we will measure a photo-induced electron-transfer reaction between two Ruthenium complexes in differing oxidation states. We will learn about the principles of diffusion-controlled electron-transfer reactions, inorganic photochemistry and photophysics, and the application of time-resolved emission spectroscopy using laser excitation to the measurement of photochemical phenomena.

An excellent introduction to the myriad applications of inorganic photochemistry is given in the review by K. Schanze and R. Schmehl in the June 1997 issue of the Journal of Chemical Education. They introduce proceedings from the August 1996 American Chemical Society Symposium, which are published in subsequent articles of the same issue. Please read a copy of this article either on the world wide web at URL: <http://jchemed.chem.wisc.edu/Journal/Issues/1997/Jun/abs633.html> or in library hard copy (Schanze, Kirk S.; Schmehl, Russell H. "Applications of Inorganic Photochemistry in the Chemical and Biological Sciences - Contemporary Developments", J. Chem. Educ. 74, 633, June 1997).

We will study a light-activated bimolecular electron-transfer reaction that can be represented by the simple equation $D^* + A \rightleftharpoons D^+ + A^-$. In principle, D and A can represent any of an enormous number of pairs of molecules that absorb light. As an electron donor, we will use an electronically excited state of the molecule $[\text{Ru}^{2+}(\text{bpy})_3]\text{Cl}_2$, where bpy (2,2'-bipyridine) is the organic bidentate ligand shown



below:

$\text{Ru}^{2+}(\text{bpy})_3$ has been the subject of intense research for the past half century. This family of molecules has major significance in bioinorganic chemistry, electron-transfer, and solar energy. The central Ru atom bears D_3 octahedral microsymmetry, as shown in Figure 1 below.

Luminescence of $\text{Ru}^{2+}(\text{bpy})_3$ was discovered nearly forty years ago. The singlet electronic transition is a charge-transfer absorption occurring at 460 nm in solution. We will use the blue edge of this broad absorption at 337 nm in order to use our available pulsed nitrogen laser. The highest occupied molecular orbital (HOMO) is localized on the Ru ion, while the lowest unoccupied molecular orbital (LUMO) is of π^* character, predominantly centered on one of the organic bipyridine ligands. The strongly allowed singlet electronic transition has a peak molar absorptivity of $\epsilon = 14,600 \text{ Lmol}^{-1}\text{cm}^{-1}$.

After excitation, a spin-conversion process occurs as the excited electron travels to a lower energy electronic state with largely triplet character. This rapid intersystem crossing (ISC) occurs within about 10^{-13} seconds, which is effectively instantaneous on the time scale of our nanosecond resolution measurement. These triplet states have a substantially longer excited state lifetime than most organic singlet excited states, e.g., one microsecond instead of 10 nanoseconds. The decay from the excited triplet state to

the ground state occurs via emission of a photon with wavelength 610 nm. The difference in energy between the 337 nm and 610 nm photons of $13,280 \text{ cm}^{-1}$ is released to the surrounding solvent molecules as heat via vibrations, rotations, and translations. A simplified photophysical energy diagram is found in Figure 2.

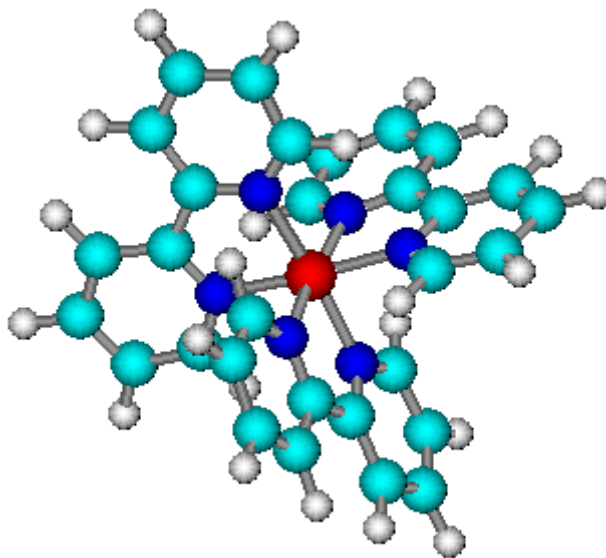


Figure 1. Ball-and-stick model of $\text{Ru}^{2+}(\text{bpy})_3$

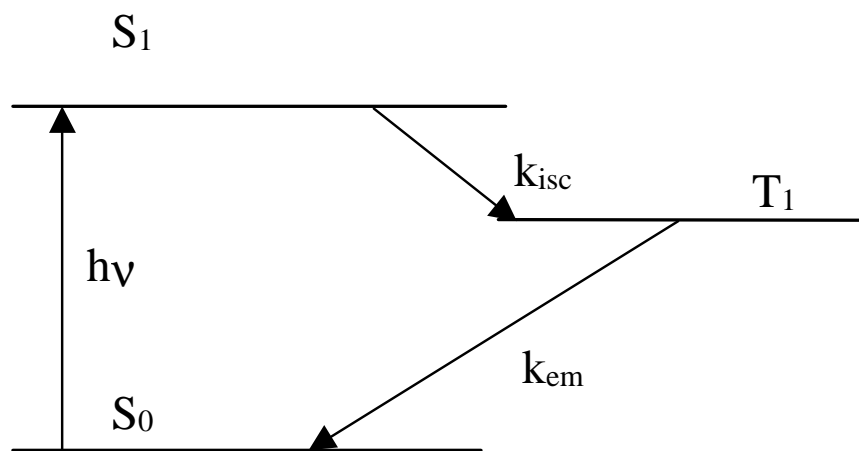
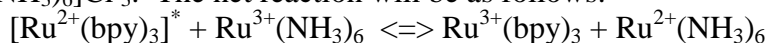


Figure 2. Simplified Energy Diagram. The observed emission is from the first triplet state (T_1) to the ground electronic state, (S_0).

Photochemistry:

We will be using the excited triplet state of $\text{Ru}^{2+}(\text{bpy})_3$ as our reagent. This excited triplet state is more reactive than the ground state both for oxidation and reduction reactions. We will be using another Ruthenium complex in the +3 valence state to oxidize the excited state of $\text{Ru}^{2+}(\text{bpy})_3$. Specifically, we will use $[\text{Ru}^{3+}(\text{NH}_3)_6]\text{Cl}_3$. The net reaction will be as follows:



The $\text{Ru}^{2+}(\text{bpy})_3$ excited state serves as the electron donor, and the $\text{Ru}^{3+}(\text{NH}_3)_6$ as the electron acceptor. Because the reaction rate for the electron transfer is very rapid, the observed rate that we will measure will be the diffusion limited rate. I.e., on the time scale of our observation, the reaction will appear to occur as quickly as the reagents can diffuse together within a small distance, or actually come into contact. By changing the concentration of the acceptor, $\text{Ru}^{3+}(\text{NH}_3)_6$, we can determine the rate of diffusion of these two ions in solution. The diffusion-limited reaction rate will be simply the slope of the observed reaction rate versus concentration. (For a discussion of diffusion-limited reactions, consult Atkins Physical Chemistry, Chapter 27).

The reaction rate for the photo-induced electron-transfer reaction will be determined directly from the excited state emission lifetimes of $\text{Ru}^{2+}(\text{bpy})_3$ measured 610 nm after exciting the solution with a 10 nanosecond pulse from the nitrogen laser at 337 nm. In the absence of quencher, the emission lifetime of the triplet metal-to-ligand charge-transfer ($^3\text{MLCT}$) state is τ_{MLCT} . In the presence of the oxidizing ruthenium complex $\text{Ru}^{3+}(\text{NH}_3)_6$, the emission lifetime will be reduced, or quenched, as the electron transfer reaction proceeds. For low concentrations, little or no quenching will be observed. As the concentrations increase towards 0.01 M, the lifetime τ_{quench} will shorten dramatically. The overall rate of reaction can be given as follows:

$$k_{\text{ET}} = (1/\tau_{\text{quench}}) - (1/\tau_{\text{MLCT}})$$

The experimental goal is to determine the diffusion-limited reaction rate from the slope of k_{ET} versus concentration of oxidative quencher, $\text{Ru}^{3+}(\text{NH}_3)_6$.

Overview of the Laser Lifetime Experiment:

You will measure the emission lifetime of a solution containing $[\text{Ru}(\text{bpy})_3]^{2+}$ in a series of solutions, all but one of which contain a quencher that will reduce the lifetime of the excited state prepared by the laser pulse. The experimental setup is shown in Figure 3. The excited state is prepared by the light from a pulsed nitrogen laser. The nitrogen laser pulse is approximately 10 nanoseconds ($1 \text{ ns} = 10^{-9}$ seconds) in duration. This is much shorter than the emission. The nitrogen laser emits light at 337 nm, which is strongly absorbed by the ruthenium complex. The light emitted from the cuvette is then collected by a lens and imaged into a grating monochromator. The wavelength resolution of the monochromator is varied by opening or closing the entrance and exit slits. The light is then detected by a photomultiplier tube, which converts the light into an electrical voltage signal. This signal is then sent into a digital oscilloscope. The input impedance of the oscilloscope is externally terminated to 50 ohms in order to minimize the RC time constant of the detection system. The RC time constant must be much smaller than the emission lifetime, or else the emission signal observed on the oscilloscope will be

distorted. The oscilloscope is triggered by a signal from a photodiode, which is aligned to detect the laser pulse. The triggering of the oscilloscope initiates the trace observed on the screen. The digital oscilloscope allows for the repetitive acquisition of data and its subsequent averaging; we can program the LT322 scope to do up to 4000 averages.

Avoiding Multiphoton Effects:

If the laser pulses are sufficiently bright so that it is possible for two or more photons to interact with an individual chromophore molecule, then multiphoton processes can occur. Multiphoton processes include ionization and dissociation of the molecule, as well as absorption to quantum states that are not directly resonant with the laser wavelength.

To estimate whether or not we can have multiphoton effects, we must satisfy the condition that the number of quanta of laser photons must be less than the number of absorbing molecules.

We can assume that our nitrogen laser is focused to a 1.0 mm diameter cylindrical beam in the sample cell, which has a path length of 10 mm. The nitrogen laser operates at a wavelength of 337 nm. From the relation $c = \lambda \nu$, one can estimate the energy per photon from $E = h \nu$. The nitrogen laser specification indicates that the laser energy is 120 $\mu\text{J}/\text{pulse}$. From these parameters, one can calculate both the illuminated volume, and the number of photons within this volume. Assume that the absorbance is 0.3 across the 10 mm cuvette path length.

To ensure that we do not have multi-photon effects, we must compare the number of photons absorbed within the cylindrical laser beam volume with the number of absorbing $\text{Ru}^{2+}(\text{bpy})_3$ complex molecules in this volume. Assume that the concentration of this solution is 10^{-4} M . From these values, calculate the number of molecules that were in the path of the 337 nm laser beam. Compare this with the number of photons absorbed.

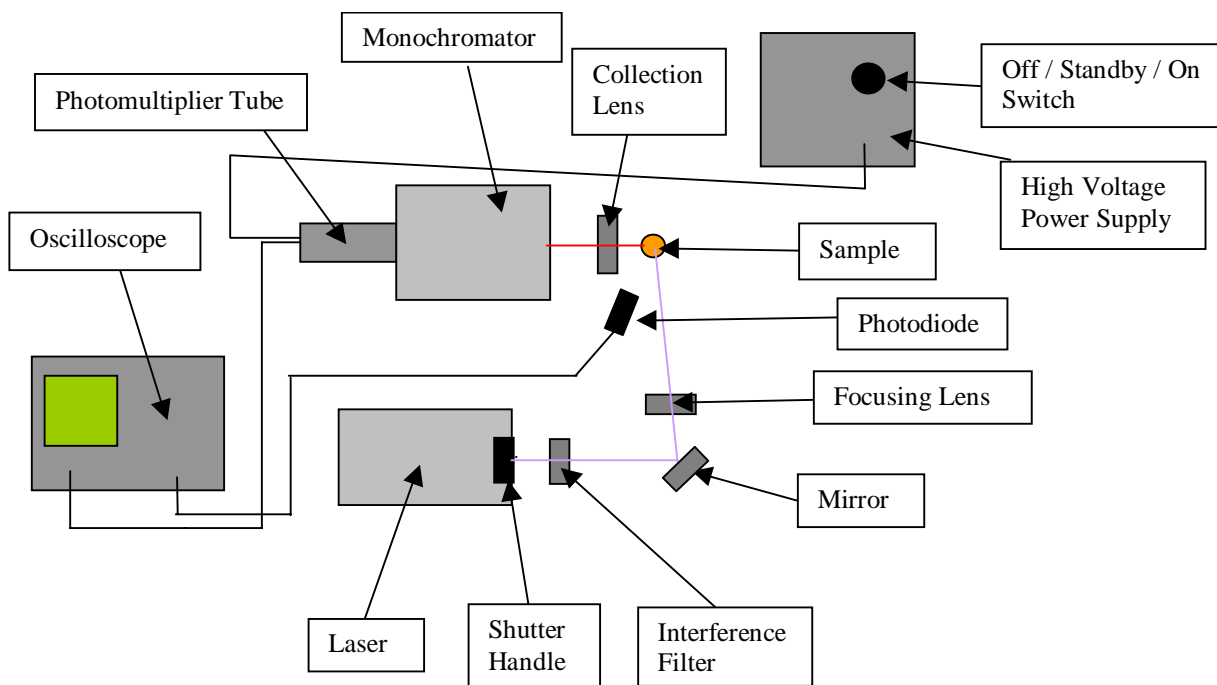


Figure 3. Experimental Diagram

**NOTE: YOU MUST WATCH THE SAFETY VIDEO
BEFORE YOU ATTEMPT THE EXPERIMENT.**

Experimental Procedure:

1. Prepare for the laser experiment by viewing the Coherent Corp. Laser Safety Training video presentation. Specific laser safety tips are appended at the bottom of this document.
2. Familiarize yourself with the laser emission lifetime setup by comparing the instrument in the lab with the diagram above. Make sure you know what each of the components does.
3. Make solutions of $\text{Ru}^{2+}(\text{bpy})_3$ with $\text{Ru}^{3+}(\text{NH}_3)_6$ quencher concentrations of 0, 3×10^{-4} M, 1×10^{-3} M, 3×10^{-3} M, and 6×10^{-3} M. The $\text{Ru}^{2+}(\text{bpy})_3$ stock solution will be designed to provide an absorbance of about 0.45 in the 1.0 cm path cuvette (i.e., about 10^{-4} M). It will be provided by your TA. Use the 2×10^{-2} M solution of quencher to make the following dilutions. The solution of quencher should be prepared by yourself. Remember that standard absorption/emission cuvettes contain at most only about 4.5 ml. You can make 50 ml 2×10^{-2} M quencher. It should be enough for your experiment.
 - a. Add 2.0 ml of $\text{Ru}^{2+}(\text{bpy})_3$ to the cuvette 1 with zero quencher, and add 2.0 ml of distilled, deionized water.
 - b. Add 2.0 ml of $\text{Ru}^{2+}(\text{bpy})_3$ to cuvette 2 with diluted quencher: dilute 3.0 ml of 2×10^{-2} M quencher solution into 97.0 ml distilled, deionized water. Add 2.0 ml of the resulting 6×10^{-4} M quencher to cuvette 2; cap; shake gently once.
 - c. Add 2.0 ml of $\text{Ru}^{2+}(\text{bpy})_3$ to cuvette 3 with diluted quencher: dilute 1.0 ml of 2×10^{-2} M quencher solution into 9.0 ml distilled, deionized water. Add 2.0 ml of the resulting 2×10^{-3} M quencher solution to cuvette 3.
 - d. Add 2.0 ml of $\text{Ru}^{2+}(\text{bpy})_3$ to cuvette 4 with diluted quencher: Add 3.0 ml of quencher solution to 7.0 ml water; add 2.0 ml of the resulting 6×10^{-3} M solution to cuvette 4.
 - e. Add 2.0 ml of $\text{Ru}^{2+}(\text{bpy})_3$ stock solution to cuvette 5. Add 6.0 ml of quencher solution to 4.0 ml water; add 2.0 ml of the resulting 1.2×10^{-2} M solution to cuvette 5.
4. Put on laser safety goggles. Close the shutter to the laser. Turn on laser with key interlock switch **while shutter remains closed**. Turn pulse repetition dial fully clockwise for maximum (20 Hz) repetition rate.
5. **Gently** turn on the photodiode circuit by sliding the switch forward to the On position.
6. Place cuvette 1 containing only the $\text{Ru}^{2+}(\text{bpy})_3$ solution into the cuvette holder.
7. Turn on the LT 322 digital oscilloscope. Ensure that the signal from the photomultiplier (detector attached to monochromator, and with high voltage input cable) goes into Channel 1.
8. Ensure that the photodiode is connected to Channel 2 of the oscilloscope.

9. Turn on the Bertan high-voltage power supply via the toggle switch located at the bottom. The setting of -750 Volts **must not be changed**. The high voltage cable must not be disconnected, for obvious safety reasons.

10. Open the laser shutter. The oscilloscope should show a brief spike trace on channel 2 from the scattered laser signal. Set the positive-going, DC-coupled trigger level to between 10 and 100 mV on this channel 2 signal. Adjust the vertical offset and variable gain for both channels 1 and 2 to fill the screen. This maximizes the signal quality available from the analog-to-digital converter at the scope inputs.

11. Obtain the emission lifetime for the zero-quencher case from the exponential decay law

$$I(t) = I(0) \exp(-t/\tau_{MLCT})$$

12. Repeat this procedure for cuvettes 2, 3, 4, and 5.

13. Obtain the diffusion limited quenching rate from the plot of rate versus concentration.

14. Shut down the photodiode, oscilloscope, high-voltage power supply, and nitrogen laser.

Error Analysis:

We would like to estimate the total error in the nearly diffusion limited rate. You will make a plot of k_{et} versus concentration. Consider the following sources of error in your data:

1. The measured intensity has an intrinsic accuracy limited by the 8-bit analog-to-digital converter of the oscilloscope. Thus, the accuracy is $1/(2^8) = 0.4\%$. The noise in the experiment has a large range, perhaps up to $\pm 10\%$. Averaging removes this. Discuss the signal to noise ratio (fluctuation from average value) observed after averaging.
2. The oscilloscope cursors allow you to estimate the error in time. Propagate the error in time into an error in lifetime τ , and hence rate k .
3. Consider the error in concentration: there is an error of 1 mg in weighing the quencher in an amount of 300 mg, or 0.33%. The pipettes used can be estimated to be good to 0.5%. Propagate these two errors to determine the error in quencher concentration. Then consider the two volumes mixed by pipette- each should have an error value from the two pipetting steps. Propagate this error.
4. Deduce a final error value.

Laboratory Report:

Consult the reference for the reaction rate and compare it with your value. The value should be found in the 1974 article by Navon and Sutin in *Inorganic Chemistry*: *Inorg. Chem.* (1974) 13, 2159.

Follow the procedure in Shoemaker, *et al.* for preparing your lab reports.

Consider the error propagation suggestions above.

Calculate whether multi-photon effects are important in this experiment. First, calculate the number of photons in the 120 μJ pulse, and separately calculate the volume subtended by an assumed cylindrical laser beam of 1.0 mm diameter and 10 mm length (in the cuvette). Assume an absorbance of 0.3 in the cuvette. The remainder of the photons are transmitted through the cuvette, the rest are absorbed. Multiply this fraction by the number of photons in the pulse.

Next, calculate the number of molecules in the sample volume, using the description given in the Multiphoton Effects section. Compare the number of photons in the sample with the number of molecules in this path. Is it likely that we can have multi-photon effects in this experiment?

Requirements for lab report:

1. Use a spreadsheet program, such as Origin, Igor, Excel, or other of your choice. Prepare graphs from your oscilloscope traces (Trace B) for cuvettes 1, 2, 3, 4, and 5. You will have saved these using the LeCroy Scope Explorer program. Plot Intensity (y axis) (Volts) versus Time (x axis).
2. Do a nonlinear least-squares fit of your data to a single-exponential function. To do this, you will need to use the following algorithm:
 - A. Find the centroid of the peak on your decay trace. The x-axis must be shifted so that this value reads zero time. Take the value observed for the peak, and subtract this value from the entire Time (x axis) column in your spreadsheet program.
 - B. There will be a non-zero baseline on your trace. You must use a fit function with a baseline added.
 - C. The fit function you will need to use is:
$$I(t) = B + I(t=0) \times \exp(-k t)$$
where B is the baseline value, $I(t=0)$ is the value at the peak of the curve, and k is the rate (in units of second^{-1}).
 - D. You will obtain rates from this analysis for each cuvette. Plot them on a graph of rate versus quencher concentration, or k vs. [Q]. The slope of this graph will give you the rate constant in units of $\text{M}^{-1}\text{s}^{-1}$.
3. Compare your obtained rate constant with the value of $2.1 \times 10^9 \text{ M}^{-1}\text{s}^{-1}$ published by Navon and Sutin in 1974, *Inorganic Chemistry*. Explain any discrepancies in detail.

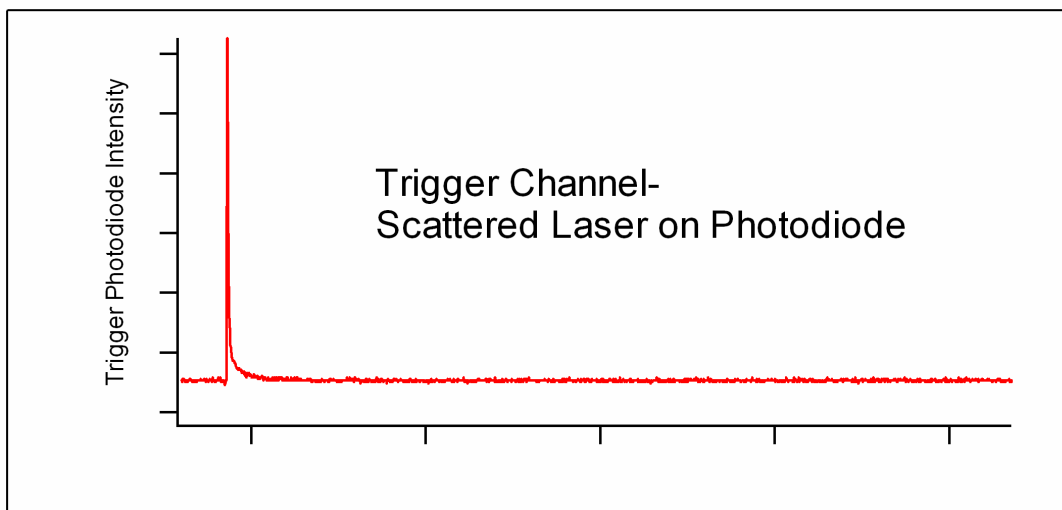
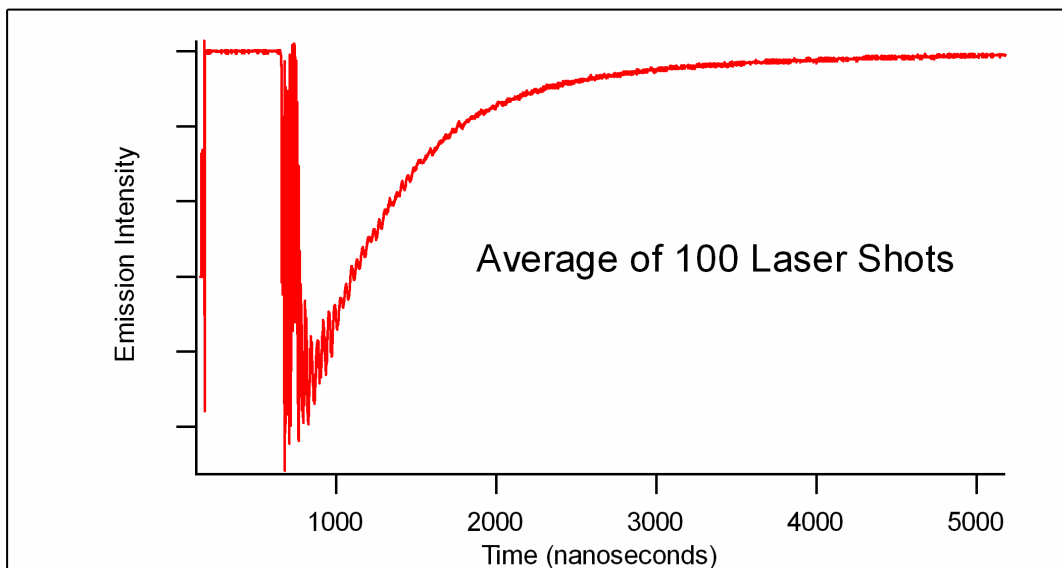
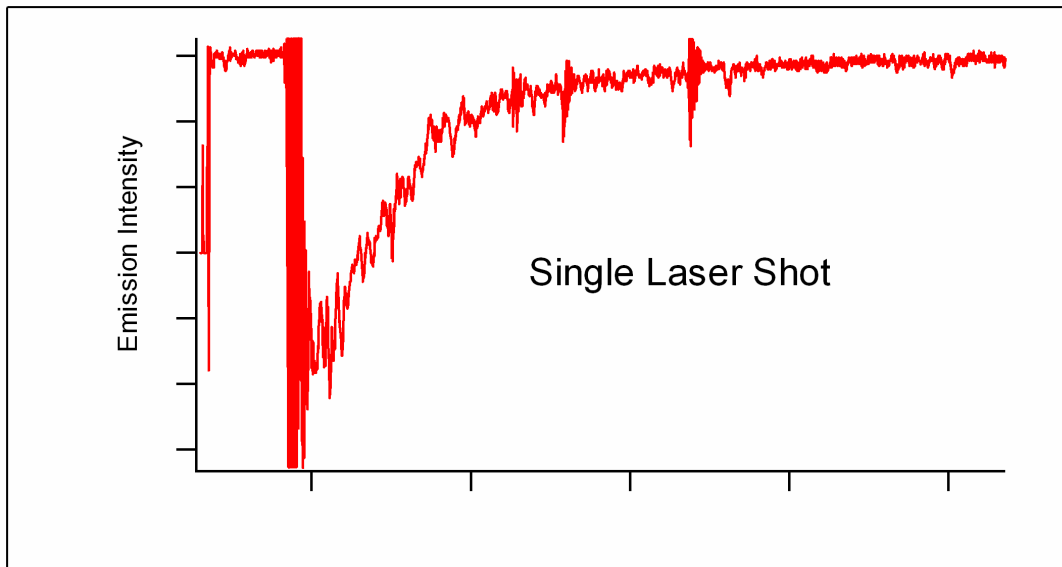


Figure 3. Sample oscilloscope traces for $\text{Ru}^{2+}(\text{bpy})_3$ in water.

Notes on LeCroy LT322 digital oscilloscope operation with the laser kinetics experiment.

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0. **WEAR GOGGLES.** Before all else, make sure that you, your lab partner, and anybody working nearby has adequate laser eye protection. Please wear the orange-lens laser goggles supplied. Neighbors working nearby in the lab must wear standard polycarbonate safety goggles; these are approved at several universities for use blocking nitrogen-laser 337 nm radiation, absorbing more than 99.99% of indirect laser light.
1. **Turn on scope.** Turn the LT 322 digital scope power on using the switch labeled "0 1" in the lower left corner of the front panel. Make sure that the photomultiplier (PMT) BNC cable is connected to Channel 1, and that the photodiode (trigger) signal is connected to Channel 2.
2. **Turn on power for photodetectors.** Ensure that the power is turned on for the photodiode (switch on top) and that the high voltage supply for the photomultiplier is turned on (set to -750 V, DC).
3. **Use pre-stored scope setups.** Press the button labeled "Panels" in the middle of the front panel controls of the LT 322 scope. The menu will appear on the right side of the screen. Choose "Recall" (***NOT SAVE!***) on the top-most button directly to the right of the on-screen menu. Next, choose one of the four setups, e.g., "Setup1". Should all of the setups be corrupted by an evil demon, ask your TA to restore the settings from the magic floppy which I have previously provided to them. Because you can always recover the original functioning settings, do not hesitate to experiment with any and all of the scope settings. Panel Setup 1 will have left the scope in a display mode with four different waveforms displayed in a vertical stack, in the Traces labeled "1", "2", "A", and "B". Panel Setup 2 will display only the averaged and inverted data in Trace B.
4. **Typical parameter settings.** Observe the on-screen settings. Typical values are given below:

Time base:	0.5 microsecond/div.
Display:	Single channel
Trigger mode:	Edge trigger on Channel 2, Positive slope, DC coupled, 100 mV trigger threshold.
Channel 1:	between 5 to 10 mV/div, DC coupled, 50 Ohm
Channel 2:	100 mV/div, DC coupled, 50 Ohm.
Zoom/Math A:	Math function "Average", 200 shots, Sum.
Zoom/Math B:	B = -A.
Trigger Mode:	Normal (instead of Auto, Stop, or Single Trigger).
Channel 1 Offset:	Manually adjust so noise baseline before trigger is near top of screen.

Trigger Position: Set so that trigger point is one division in from left edge of screen.

5. **Time window.** For $\text{Ru}^{2+}(\text{bpy})_3$ aqueous solution with no added quencher, we want to begin with a 5 microsecond time window, perfectly appropriate for measuring the full decay trace for a 1 microsecond 1/e time constant. The full screen has ten vertical divisions, so we set the time base to 0.5 microseconds/division. As we add increasing concentrations of quencher, the lifetime will decrease from 10^{-6} s to about 5×10^{-8} s, so you will need to change the time per division to 0.2, then 0.1 microseconds/div.
6. **Begin measurements.** When your sample is ready, laser safety goggles firmly in place, make sure that the laser shutter is closed. Place the fluorescence cuvette carefully in the cuvette holder, being careful not to put fingerprints on the optical surface of the cell. Open the laser shutter by carefully pulling upwards. Observe that the Channel 1 signal is present.
7. **Optimize variable gain and vertical offset.** You must manually adjust the vertical scale to fill, *but not overflow* the vertical range on the screen. This will require that you press Channel 1 button (far right of scope), and press the "Gain: variable" button. Then adjust the gain between 5.0 and 10.0 mV/div using the Vertical Scale knob, while maintaining the zero-Volt baseline near the top of the screen using the "Offset" knob.
8. **Averaging into Zoom/Math channels A-D.** Remember that when you change any setting on the oscilloscope, the averaging on Channel A (or anywhere else, for that matter) will automatically restart at zero. Also, a change in any setting may trigger an automatic recalibration event in the scope- it will stop responding for about five seconds, and state "recalibrating" at the top of the screen, and resume normal operation shortly. To restart the averaging in Channel A from zero, press the "Clear Sweeps" button.
9. **Waveform Storage.** When you have acquired an appropriate looking trace in channel B, you will now need to save your data. You should use the Scope Explorer program on the PC to which the LT322 scope is interfaced, with the directions below. Should the computer be otherwise occupied, as an alternative, you can store your data directly to a floppy diskette.

Download of averaged data from LeCroy LT322 digital scope using Scope Explorer.

- A. **Launch Scope Explorer.** Use the personal computer (Dell Gx1) to which the LT 322 scope is interfaced via the GPIB cable (rear of scope to PCI card in rear of computer). From the Start menu on computer, launch the program called *Scope Explorer*.
- B. **Resize the screen:** using the *Maximize* button at the top right of the window.
- C. **Open a Display Window.** Use either the "Display" box on the menu bar, or use the File => New Display Image command.
- D. **Open Trace List.** Use either the "Trace" button on the menu bar, or use the File => New Trace List command. Select the averaged trace that you desire to save (in this case, Trace B) by a single left-click on the mouse.

- E. Get Trace B data. Go to the *Trace* menu, => Get in ASCII Form. Select the *Byte* radio button under *Size*, and click *OK*.
- F. Choose the ASCII Trace Format. Select the *Time and Amplitude* radio button, with the *Space* radio button under *Delimiter*, then click *OK*.
- G. Save the data to a hard disk file. In the *Save As* dialog box, select the directory with your TAs name, e.g., C:\pchemlab*YourTA'sName*\ and choose a sensible filename, such as TB_xyz where xyz are your initials.
- H. Go on to the next sample.

Download of LT322 data to a floppy diskette.

- a. Insert an IBM formatted 3.5" floppy diskette into the drive at top right, top side up.
- b. Press the "*Wave Storage*" button on the bottom center region of the scope. On the right-hand-side of the screen, (under the *W'form Menu* listing) choose the *Store Waveform* item.
- c. At the bottom right of screen, select *Store -> B* and *To -> Flpy* to store Trace B to the floppy diskette. Make your selections by pressing the buttons or moving the knobs to the correct selection. (The default will normally be "Store 1 to M1"...)
- d. Choose "Ascii" format (*NOT BINARY!*) in the top menu button.
- e. Now press "*Do Store B -> flpy*".
- f. Have your notebook and pen ready to write down the filename next to your experimental conditions: an example might be "STB003.txt". If the filename begins with anything else besides "STB...", (e.g., "ST2..."), you have made a mistake. Go back and correctly save your data before proceeding.
- g. Remove the floppy diskette, and load data into computer using a spreadsheet program like Excel, or better.

LASER SAFETY

- A. The N₂ laser is Class IV, the most hazardous class.
- B. The laser pulse parameters are: up to 200 microJoules/pulse, 20 pulses/sec, all at 337 nanometers. This is UV-B radiation, and direct exposure over time can cause sunburn, and likely skin cancer.
- C. Class IV laser radiation can cause permanent damage to corneas, lenses, and retinas from either direct or indirect exposure.
- D. Students must wear appropriate laser goggles or polycarbonate safety glasses (that have higher than Optical Density (absorbance) of 4.0 at 337 nm) such as the four sets of Uvex glasses provided. Neighboring students must also wear polycarbonate lenses as well.
- E. The laser shutter must remain closed at all times except when data is being collected.
- F. Students should wear no jewelry that could in any way intersect the laser beam. Examples of taboo jewelry include rings, wrist-watches, or dangling earrings, necklaces, etc.
- G. The shutter should be closed while cuvettes are being added or removed from the beam path.
- H. Students should never place their eyes anywhere near the height of the laser output, as the brightest direct reflections will occur in this plane.
- I. The nitrogen laser output is invisible to us. Violet/blue spots observed (e.g., from the business card) are visible fluorescence.
- J. The cover to the laser must never be removed by students under any circumstances.