CHEMISTRY 260

Spring 2006

Book required : "Spectrometric Identification of Organic Compounds," 7th ed., by Silverstein and Webster (an earlier edition will suffice)

You might want a copy of an instrumental analysis textbook – any will do, and there are a couple on library reserve.

Goals:

A. For the following techniques, which we will concentrate on, you should understand the theory, understand how the instrument works, and be able to interpret the data:

- Ultraviolet and visible spectroscopy
- Infrared spectroscopy (IR)
- Nuclear magnetic resonance (NMR) spectrometry
- Mass spectrometry (MS)
- Atomic absorption spectroscopy (AA)
- Gas chromatography (GC)
- Thin-layer chromatography (TLC)
- High-performance liquid chromatography (HPLC)

B. You should know what the following techniques are and what they are used for:

- Fluorescence spectroscopy
- X-ray and electron spectroscopy
- Polarimetry
- Thermal analysis
- Nuclear methods of analysis
- Electrochemistry

C. You should also be able to:

- Identify an organic compound from its spectra
- Understand how statistics is used in analytical chemistry and interpret a statistical analysis of data
- Write a scientific report

There will be two exams. Exam I will cover spectroscopy; it will be given the week after spring break (March 19-23). There will be no lab that week; the exam may be taken anytime that week, including the regular lab time. It may be taken in one part or two parts. Exam II will cover only those topics covered since Exam I; it may be taken any time the last full week of classes up through reading day (Apr. 21-30). It may also be taken as a whole or in two parts. Although writing won't be strictly graded on the exams, if your writing is very poor, your exam grade may be lowered.

There will be a spectroscopy problem set assigned; this is to be treated like a take-home exam. The assignment will be given before spring break and will be due Friday, March 23 (the Friday after spring break). You may use only your book(s) and lecture notes for the problem set. This is not a writing assignment, so you don't have to compose a report.

There will be a final project – each lab will choose one of the instruments and prepare a video illustrating the theory, the operation, and what the results are used for. This will not be graded, but is necessary for passing the course.

The Honor Code applies to the exams, the problem set, and the lab reports. It is permissible to discuss lab reports prior to writing them up, but the report itself must be your work alone (using critiques from other students; see below). The lab report for Experiment 3 (Report II) will be written jointly by the group of students who worked together on the experiment. These students will turn in one report and all students in the group will receive the same grade on the report. See the Honor Code handout for more information.

Grading:	Exam I	25 %
	Exam II	25 %
	Problem set	10 %
	Lab	40 %

As this is a writing-intensive course, conferencing and revision are important components. You should turn in a draft of your first two lab reports for me to look over before you turn in these reports for a grade; I will meet with you after I've critiqued your draft and you can revise it. An outline of what should be included in each report will be posted to our class conference.

SCHEDULE

Date	Experiment	Reports
Feb. 1	1 (UV-VIS)	
Feb. 8	2 (UV-VIS)	>Draft of theory/instrumentation parts of **Report I** (Exp. 1-2) due
Feb. 15	3 (IR-NMR-MS)	>Draft of procedure/ results/discussion parts of **Report 1** (Exp. 1-2) due
Feb. 22	---	**>Report 1** (Exp. 1-2) due >Draft of theory/ instrumentation parts of **Report II** (Exp. 3) due
Mar. 1	---	>Draft of procedure/results/discussion parts of **Report II** (Exp. 3) due
Mar. 8	4 (AA)	> **Report II** (Exp. 3) due Friday, Mar. 9, 4:00
Mar. 15	--- (spring break)	
Mar. 22	---	>Exam I this week (finish by Friday, Mar. 23, 4:00) >Problem set due by Friday, Mar. 23, 4:00
Mar. 29	5 (GC)	>**Report III** (Exp. 4) due
Apr. 5	6 (TLC)	>**Report IV** (Exp. 5) due
Apr. 12	7 (HPLC)	
Apr. 19	---	>**Report V** (Exp. 6-7) due
Apr. 26	---	>**Report VI** (Statistics) due >Exam II this week (finish by Wednesday, May 2, 4:00) >Videos due by Wednesday, May 2, 4:00

(Reports are due at the beginning of lab time on Thursday unless otherwise specified)

CHEMISTRY 260 LAB

1. Before the lab. You should come into the lab with some knowledge of the experiment to be performed. This means having read the experiment, performed any preliminary calculations necessary, and reviewed your class notes and the relevant textbook sections.

2. During the lab. The experiment must be performed and completed during the lab period. Work efficiently and make good use of your time. You are expected to be present in the lab until the experiment is finished and to participate in all parts. Most of the glassware and chemicals needed can be obtained from the stockroom. Chemical preparation will be carried out in the prep room or room 203; use the hood when necessary. The instruments are expensive and must be handled carefully. If an instrument is broken, this not only involves the expense of repairing it, but may also entail down time before service can be arranged.

3. After the lab. Before leaving the lab, all equipment must be cleaned and put away. Any equipment obtained from the stockroom must be returned.

4. Safety. Proper safety precautions must be observed at all times. This means wearing approved safety glasses, no eating or drinking in the lab or instrument room, wearing proper protective clothing (e.g., no sandals), not wearing contact lenses, and no unauthorized experimentation.

5. Reports. All data collected in lab should be recorded in a notebook. All people in a lab will work on the same experiment and instrument. This means everybody must participate in each part of the procedure -- preparation, instrument operation, and clean-up. The report itself is to be your work alone -- any collaboration is a violation of the Honor Code. (For Experiment 3/Report II, two students will work together to prepare a single report; both students will receive the same grade.) In most cases, there will be only one original record of the data obtained directly from the instrument. If so, one person should turn in the original with his or her report and the others should mention whose report includes this record. If desired, you may copy or photocopy the data for your own report.

6. Report style. A written report is required for each technique investigated. The report is due at the beginning of the lab as noted on the schedule. The report should include:

a. Your name and the names of the others who worked on the experiment.
b. Date of experiment and date of report.
c. Number and title of experiment.
d. Technique and theory.
e. Description of apparatus used.
f. Brief description of the problem investigated.
g. Data (normally organized into tables).
h. Graphs, equations, diagrams, etc., where appropriate.
i. Results
j. Discussion and conclusions.

Reports must be written in proper scientific English -- using passive voice and past tense where appropriate. Reports should be word processed and double-spaced; they must be neat and legible with correct spelling, grammar, and punctuation. Normally, you should avoid writing anything in -- use the word processor instead.

7. Grading. Reports will be graded on a 100-point basis. You will be graded on the report itself (style, organization, completeness), your data, your results. The most important aspect is your writing and your explanation of theory and instrumentation. Some of the grading will, by the nature of the reports, be subjective.

Your lab grade will be computed by averaging the report grades and an evaluation of your performance in lab (participation, handling of equipment, clean-up, safety procedures).

EXPERIMENT 1

VISIBLE SPECTROSCOPY

Chemicals: 0.2-M $NiCl_2$

Apparatus: Shimazdu 1601 spectrophotometer, plastic cuvettes

Procedure:

Prepare a series of dilutions of the nickel solution: 0.1 M, 0.05 M, and 0.025 M.

Select "Spectrum" mode on the instrument. Place the 0.2-M nickel solution in a cuvette in the front cell holder and an empty cuvette in the rear holder. Select absorbance for the Y-axis with a maximum of 1.00. Obtain the spectrum of the nickel solution by scanning from 700 nm to 400 nm. Determine the wavelength of maximum absorbance using the zoom and peak functions.

Select "Photometric" mode on the instrument and go to the wavelength you found. Measure the absorbance of each of the nickel solutions, including the unknown. From the standard solutions, construct a Beer's Law curve of absorbance vs. concentration. If a point appears to be too far off the line, discard it.

Using the standard curve, determine the concentration of the unknown nickel solution.

No report is due until you finish Experiment 2. In your report for this experiment, you will need to include the spectrum of nickel, the wavelength selected, the Beer's Law plot, all the absorbance readings, and the concentration of the unknown.

EXPERIMENT 2

SIMULTANEOUS MULTI-COMPONENT ANALYSIS USING UV SPECTROSCOPY

Chemicals: pheniramine maleate (PAM), 80 μg/mL in 0.010-M HCl
phenylephrine hydrochloride (PEH), 200 μg/mL in 0.010-M HCl
0.010-M HCl
commercial Dristan® sample

Apparatus: Shimadzu 1601 spectrophotometer, quartz cuvettes.

Procedure:

Prepare a series of dilutions using 0.010-M HCl: PAM -- 40, 20, 10, and 5 μg/mL; PEH -- 100, 50, 25, and 12.5 μg/mL Also, dilute the Dristan® sample 1:200 with the 0.010-M HCl (this means 1 mL diluted to 200 mL total volume).

Select "Spectrum" mode on the instrument. Scan each of the standard substances. Select two wavelengths to use where each substance is at or near its maximum.

Select "Photometric" mode on the instrument. Measure the absorbances of all the solutions at each of the two wavelengths (4 solutions X 2 substances X 2 wavelengths); also measure the absorbance of the diluted Dristan® solution at each wavelength. For each substance, use Excel to construct a Beer's Law curve at each wavelength (2 substances X 2 wavelengths); determine the absorptivity (slope) of each curve.

Solve for the concentrations of the unknown solution using the matrix method in Excel.

Once you have the concentrations in the diluted Dristan®, you need to calculate what the concentrations were before dilution and then convert from μg/mL to %:

Multiply by 200 to account for the dilution. Divide by 10^6 to convert from μg/mL to g/mL (since 1 mL of dilute aqueous solution has a mass of 1 g). Multiply by 100 to convert to %.

In the report, include all spectra, Beer's Law curves, and absorbance readings. Show how you solved the equations using matrices. Give the concentrations in the original Dristan®. Discuss the theory of uv-visible absorption and the instrument.

EXPERIMENT 3

QUALITATIVE ANALYSIS

Chemicals: unknown organic compound
TMS

Apparatus: Anasazi EFT-60 NMR spectrometer, NMR tubes
Perkin-Elmer Paragon 500 IR spectrometer, salt plates

Procedure:

Your group of students will be given 2 pure, liquid, organic unknowns; they will contain only carbon, hydrogen, oxygen, and/or nitrogen.

Fill an NMR tube about 1/3 full with your unknown; add an equal amount of TMS. Obtain the NMR spectrum, including the integration trace. Clean out the NMR tube with acetone.

Place a drop of the sample between two salt plates. Obtain an infrared spectrum. Clean the salt plates with chloroform.

You will be provided with mass spectral data for your unknowns. You will be provided with carbon-13 NMR data for one of your unknowns.

Determine the identity of your unknowns, including structure and proper name. Each peak in the NMR spectra must be assigned and the shift and area explained. The major peaks in the IR and mass spectra must also be assigned. After you have identified the unknown, go back and explain how it would (or would not) be expected to give the observed spectra.

Predict what the C-13 NMR spectrum of your other unknown would look like.

This is a two-week experiment, with the report due after the second week.

In your report, include a discussion of the theory and instrumentation of scanning proton NMR spectrometry and IR spectroscopy. It is not necessary to discuss the instrumentation of C-13 NMR or mass spectrometry since you did not use these instruments, but you should discuss the theory. Be sure that once you have identified your unknowns, you compare the shift, area, and multiplicity of each NMR peak with the predicted values and that you assign the major mass and IR spectral peaks.

Each group of students will work together to identify their unknowns and to write up the report. Only one report will be accepted from the group and all students in the group will receive the same grade.

EXPERIMENT 4

ATOMIC ABSORPTION SPECTROSCOPY

Chemicals: lead nitrate or copper nitrate

Apparatus: Buck 200A atomic absorption spectrometer

Procedure:

Lead or copper may be used. By diluting the standard provided, prepare standard of 1, 2, and 5 ppm.

Prepare standard addition solutions. Mix equal quantities of your unknown with each of the standards and deionized water (0 ppm), giving 5 standard addition solutions (e.g., one such solution might be 1 mL of unknown + 1 mL of 1 ppm).

Locate the wavelength (283.3 nm for lead, 324.7 nm for copper). Set zero while aspirating deionized water. Aspirate the standards, unknown, and standard addition solutions and record the absorbances.

Construct a standard curve and determine the concentration of the unknown. Also construct a plot of the standard addition solutions (absorbance vs. ppm added) and determine the concentration of the unknown from this graph.

In your report, include the 2 plots, absorbances. and the concentration of the unknown from the 2 methods. Discuss the theory and instrumentation.

EXPERIMENT 5

GAS CHROMATOGRAPHY

Chemicals: heptane, tetrahydrofuran (THF), 2-butanone, n-propanol

Apparatus: Gow-Mac gas chromatograph with DC-stationary phase packed column and thermal conductivity detector
computer data acquisition system

Procedure:

The temperature and flow rate will be set. Inject 1 μL of the mixture, along with 5 μL of air. Inject 1 μL of each pure substance, plus air. For your report, you will need to decide which peak in the mixture corresponds to which substance by matching the retention times. From the areas in the mixture, determine the volume in μL of each substance and calculate the composition of the mixture.

In your report, include the retention times and areas of the standards and the composition of the mixture. Discuss the theory and instrumentation. Calculate the number of theoretical plates and HETP using your last peak, and α (separation) and R (resolution) using your last two peaks.

EXPERIMENT 6

THIN-LAYER CHROMATOGRAPHY

Chemicals: vitamin standards; may include ascorbic acid (vitamin C), nicotinamide (a B vitamin), vitamin A (as the acetate or aldehyde), and tocopherol (vitamin E); a mixture of the vitamin standards

developing solution (ethyl acetate/ethanol 2:1 or 3:1)

Apparatus: TLC plates, polyester coated with silica gel
TLC developing tank
uv lamp

Procedure:

Prepare a solution of each vitamin as 0.01 g/mL in ethanol.

Place the developing solution in the tank and allow the atmosphere to become saturated.

Mark a starting point on a TLC plate with a pencil. Spot the plate with the solutions, including the mixture, using a capillary tube. Allow the solvent to dry and add a second spot on top of the first. When the solvent has dried again, place the plate in the developing solution in the tank. Allow the solvent to move as far as it can, remove the plate, mark the solvent front with pencil, and allow the solvent to dry. View the plate under a uv lamp. The plates have been treated with a fluorescent material, so the vitamins will appear as dark spots against a bright background. Circle each of the spots and calculate R_f values.

Look up the structures of the vitamins. Discuss the differences in R_f values in terms of the polarities of the vitamins and the mobile phase. (The stationary phase, silica gel, is quite polar; the mobile phase is only slightly polar.). Discuss how the polarity of a vitamin determines whether it is fat-soluble or water-soluble.

No report is necessary until Experiment 7 is finished.

EXPERIMENT 7

HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY

Chemicals: 1.0 % toluene in methanol
0.01 % benzaldehyde in methanol
0.2% N,N-diethyl-m-toluamide (DET) in methanol
methanol, water

Apparatus: ISCO 2361 HPLC
chart recorder

Procedure:

Different concentrations are necessary because benzaldehyde absorbs strongly at 254 nm, DET absorbs moderately, and toluene absorbs weakly. Run the mixture of all 3 at a flow rate of 1 mL/min using a mobile phase of 70 % methanol/water.

Run each individual standard. From the retention times, identify each component in the mixture. Determine the % of each component in the mixture:

$$\frac{\text{area of standard}}{\text{area of unknown}} = \frac{\text{\% of standard}}{\text{\% of unknown}}$$

Note that the percentages will not add to 100%; they should be close to the percentages of the standards.

In your report, discuss liquid chromatography in general and specifically the two types you used, TLC and HPLC. Discuss the instrumentation used for HPLC.

STATISTICS

EVALUATION OF DATA

Using the techniques discussed in class, two sets of data are to be analyzed. The first set of data is to be used to fit a first-order (linear) model. You should report the coefficients, their significance levels, the 5 sums of squares (corrected, regression, residual, lack of fit, and pure error), and the significances of regression and lack of fit.

The second set of data is to be used to fit both a first-order and a second-order (quadratic) model. Again, all of the information requested for the first data set should be reported for each mode. Also, include an evaluation as to which model fits the data better, and discuss the criteria you used to make that decision.

DATA SET 1

pH	Absorbance
2.0	0.212
2.0	0.200
3.0	0.319
3.0	0.328
4.0	0.411
4.0	0.400
5.0	0.515
5.0	0.500

DATA SET 2

pH	Temp.	Absorbance
1.0	10.0	0.288
1.0	40.0	0.462
1.0	70.0	0.510
1.0	100.0	0.432
4.0	10.0	0.429
4.0	40.0	0.533
4.0	70.0	0.507
4.0	100.0	0.432
7.0	10.0	0.514
7.0	40.0	0.542
7.0	70.0	0.440
7.0	100.0	0.207
10.0	10.0	0.537
10.0	40.0	0.488
10.0	70.0	0.310
10.0	100.0	0.002
4.0	40.0	0.537
4.0	70.0	0.510
7.0	40.0	0.534
7.0	70.0	0.425