Ch 362 Instrumental Analysis I (Fall 2022) – Lecture Schedule & Syllabus

Instructor: Prof. Athula B. Attygalle (aattygal@stevens.edu); McLean 312;

Teaching Assistants: Idris Junaid (ijunaid@stevens.edu), Oshane Annon (oannon@stevens.edu)

COURSE DESCRIPTION from Catalog

Theoretical and experimental approach to spectroscopy and chromatography. Includes ultraviolet, visible and infrared absorption by molecules, emission spectroscopy, nuclear magnetic resonance, mass spectroscopy and gas-liquid and high-performance chromatography. Prerequisites: **CH 116**, **CH 118**

Educational Aims

- To deliver a course that will provide an intellectually challenging learning experience
- To encourage student enthusiasm and curiosity.
- To develop in students a range of general laboratory skills that will help them to find gainful employment in chemistry-based industry.

Textbook: *Quantitative Chemical Analysis*, Tenth Edition| 2020, Daniel C. Harris; Charles A. Lucy. (McMillan). ISBN:9781319164300
Format: 3 lectures/week, M, W, and F, 9.00 to 9.50 am (Kidde 360). One laboratory session/week (McLean 429; M 2.00-5.50 PM; T 2.00-5.50 PM; F 2.00-5.50 PM); a written pre-laboratory report should be submitted electronically two days before each laboratory session. A hardcopy final lab report should be submitted a week after the experiment. Expect several spot prelab and post-lab quizzes.
Oral Presentation: One, at the end of term. A 15-min oral presentation using PowerPoint.
Attendance: You must be present at all lectures and laboratory sessions. (**Unexcused absences can result in a lowered final grade; all laboratory experiments must be completed**).

Lecture Schedule, MWF
Session: Normal Academic Term 09-01-22 to 12-14-22

DAY	BEGINS	ENDS	BUILDING	ROOM
MWF	09:00 AM	09:50 AM	Kidde	360

Laboratory Schedule, MTF

DAY	BEGINS	ENDS	BUILDING	ROOM
Monday (362-LA)	2:00 PM	5:50 AM	McLean	429
Tuesday (362-LB)	2:00 PM	5:50 AM	McLean	429
Thurday (362-LA)	11:00 AM	2:50 PM	McLean	429

CANVAS: A website for this course is available on https://mystevens.stevens.edu/. You can upload your prelabs and assignments on Canvas.

ACADEMIC INTEGRITY

Undergraduate Honor System

Enrollment into the undergraduate class of Stevens Institute of Technology signifies a student's commitment to the Honor System. Accordingly, the provisions of the Stevens Honor System apply to all undergraduate students in coursework and Honor Board proceedings. It is the responsibility of each student to become acquainted with and to uphold the ideals set forth in the Honor System Constitution. More information about the Honor System including the constitution, bylaws, investigative procedures, and the penalty matrix can be found online at http://web.stevens.edu/honor/

The following pledge shall be written in full and signed by every student on all submitted work (including, but not limited to, homework, projects, lab reports, code, quizzes and exams) that is assigned by the course instructor. No work shall be graded unless the pledge is written in full and signed.

"*I pledge my honor that I have abided by the Stevens Honor System*."

Reporting Honor System Violations

Students who believe a violation of the Honor System has been committed should report it within ten business days of the suspected violation. Students have the option to remain anonymous and can report violations online at www.stevens.edu/honor.

INCLUSIVITY STATEMENT

Stevens Institute of Technology believes that diversity and inclusiveness are essential to excellence in education and innovation. Our community represents a rich variety of backgrounds, experiences, demographics and perspectives and Stevens is committed to fostering a learning environment where every individual is respected and engaged. To facilitate a dynamic and inclusive educational experience, we ask all members of the community to:

- be open to the perspectives of others

- appreciate the uniqueness their colleagues
- take advantage of the opportunity to learn from each other
- exchange experiences, values and beliefs
- communicate in a respectful manner
- be aware of individuals who are marginalized and involve them
- keep confidential discussions private

COURSE SCHEDULE (any changes will be posted on Canvas)

	Date	Topic	Chapter. Sec	Reading homework* (sections refer to Harris Ninth Edition)
1	09/02	Introduction to analytical procedures. Safety. Maintenance of a Laboratory Notebook. Grading Policies	1, 2 (Harris 8th Edition) 1, 2 (Harris 9th Edition)	Revise; Concentrations, molarity, periodic table, writing correct formulas and solubility and nomenclature of inorganic compounds. 0-2 (General steps in a chemical analysis), 1-1 (SI Units), 1-2 (Chemical Concentrations), 1-3 (Preparing solutions), 2-1 (Safety), 2-2 (Lab note book), 2-3 (Analytical Balance).
2	09/05	Labor Day. No class		
3	09/07	Measurements and errors. Use of Excel	2, 3, 4 (Harris 8th Edition) 2,3 (Harris 9th Edition)	2-3 (analytical Balance), 2-4 (burets), 2-5 (Volumetric flasks), 2-6 (Pipets), 2-10 (Excel), 2-11 (Graphing; appendix A; page AP1), 3-1 (Significant Figures), 3-2 (Significant Figures in Arithmetic), 4-1 (Statistics, Gaussian distribution).
4	09/09	Volumetric methods, titrations	10 (Titrations)	10-1 (strong base with strong acid; page 206), 10-2, 10-3, 10-6 (indicators).
5	09/12	Redox titrations	15 (Redox Titrations)	15-1 (redox titration curves), 15-2 (end point), 15-4, 15-5, 15-6 (dichromate), 15-7 (iodine). Appendix D (AP 5- AP7)
6	09/14	Fundamentals of spectroscopy	17 (Harris 8th Edition)	18-1 (light; page 433), 17-2 (absorption), 17-3, 17-4 (Beer's Law),

			18 (Harris 9th Edition)	17-6 (electronic states), 19-1 (light sources).
7	09/16	Atomic spectroscopy	20	20-1(atomic spectroscopy), 20-2, 20-4,
8	09/19	Atomic absorption and emission / Quiz 1	19	20-4 (instrumentation).
9	09/21	Ultraviolet and visible spectroscopy	18, 19, 20	18-5, 19-1(light sources)
10	09/16	Ultraviolet and visible spectroscopy	18, 19, 20	18-5, 19-1(light sources)
11	09/19	UV and visible experimental procedures	18, 19, 20	17-3 (Measuring absorbance)
12	09/21	Absorption of light.	17	17-1(light), 17-2, **17-4**, 17-6 (electronic states)
13	09/23	Fluorescence	17	17-6 (Fluorescence), 17-7
14	09/26	Fluorescence	17	17-6 (Fluorescence), 17-7
15	09/28	Vibrational modes Infrared Spectroscopy	19	19-4, 19-5 (FT-IR) 19-4 (sensors; refraction, ATR), 19-5 (FT-IR)
16	09/30	Interferometry, FTIR	19	19-5 (FT-IR)
17	10/03	IR techniques, Attenuated Total Reflectance (ATR)	19	19-4
18	10/05	Fluorescence	17	17-6 (Fluorescence), 17-7
19	10/07	Fluorescence	17	17-6 (Fluorescence), 17-7
20	10/10	Recess, No Class		
21	10/11 (Monday Schedule)	Quiz 2		
22	10/12	Polarimetry		Lab Instructions manual; http://en.wikipedia.org/wiki/Polarimetry
23	10/14	Analytical Separations	22 (analytical separations)	22-1 (solvent extraction)
24	10/17	Chromatography theory & techniques	22	22-1, 22-2 (chromatography), 22-3 (retention times)

25	10/19	Chromatography theory & techniques	22	22-1, 22-2 (chromatography), 22-3 (retention times)
26	10/21	Preliminary Exam		
27	10/24	Gas chromatography (GC)/	23 (GC)	23-1 (separation process)
28	10/26	GC phases & columns	23	23-1 (separation process)
29	10/28	GC injectors	23	23-2 (sample injection), 23-4 (sample preparation)
30	10/31	GC detectors	23	23-3 (detectors)
31	11/02	Quantification	24	24-1(Chromatographic Process)
32	11/04	Liquid Chromatography	24 (HPLC)	24-1, 24-2
33	11/07	HPLC	24	24-1 (Chromatographic Process)
34	11/09	Injectors & detectors	24	24-2 (pumps, valves, detection)
35	11/11	Calibration curves and Quantification	24	24-3 (method development)
36	11/14	Overview of chromatographic process/Modern HPLC methods	25	24-4 (Gradient Separations)
37	11/16	Overview of chromatographic process/Modern HPLC methods	25	24-4 (Gradient Separations)
38	11/18	Quiz 3		
39	11/21	Overview of chromatographic process/Modern HPLC methods	25	24-4 (Gradient Separations)
40	11/23	UPLC		*www.waters.com/**uplc***
41	11/25	Affinity and Size exclusion chromatography	25	25-3 (molecular exclusion)
	11/27	Thanksgiving		
	11/29	Thanksgiving		
42	12/02	Revision		
43	12/04	Revision		
44	12/06	Revision		

45	12/?	Final Exam	8 am-12 noon?	Room?

*Reading assignments

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Ch 362 Instrumental Analysis I

Stevens Institute of Technology

(3-4-4)

Course Description:.
Experimental approach to spectroscopy. Topics include Fourier Transform infrared spectroscopy, ultraviolet, visible and fluorescence measurements, atomic absorption spectroscopy, and nuclear magnetic resonance spectroscopy.

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Lectures: Grading Scheme (Total 750 pts).

- Five random spot quizzes will be given during the class; the worst quiz score will be dropped and the rest will be summed for the final grading.

Quiz # 1	100 pts
Quiz # 2	100 pts
Quiz # 3	100 pts
Homework	200 pts
Spot Quizzes	100 pts
Total (Quizzes)	500 pts
Prelim	100 pts
Final Exam	150 pts
TOTAL	**750 pts**

Laboratory work: Grading Scheme (Total 750 pts). Seven Units.

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Ch 362 Instrumental Analysis I
Laboratory Experiments

Unit 0: Strong base/strong acid titration

Volumetric Redox Titration

Unit 1: Ultraviolet-Visible Spectroscopy

Experiment 1. Determination of Ascorbic Acid in Vitamin C Tablets.

Unit 2: Fourier Transform-Infrared Spectroscopy

Experiment 1. Characterization of Organic Polymers as Thin Films by FT-IR

Experiment 2. Characterization of Organic Compounds as Solutions by FT-IR

Experiment 3. Quantitative Determination of Methyl Ethyl Ketone

Unit 3: Spectrofluorometry

Experiment 1. Determination of Quinine in Tonic Water by Spectrofluorometry

Unit 4: Polarimetry

Experiment 1. Quantitative Determination of Sucrose by Polarimetry

Unit 5: Atomic Absorption Spectrometry

Experiment 1. Determination of Copper by Ordinary Linear Calibration Method

Experiment 2. Analysis of Copper by the Standard Addition Method

Experiment 3: Determination of Percentage Copper in an Alloy by Ordinary Linear Calibration Method

Unit 6: Gas Chromatography

Experiment 1. Gas-liquid chromatographic Separation of a Hydrocarbon Mixture

Experiment 2. Alcohol determination

Unit 7: High Pressure Liquid Chromatography

Experiment 1. HPLC Separation of a Mixture of Compounds

Experiment 2. HPLC Determination of Caffeine in Beverages

Goals:

To provide students basic analytical skills to:

1. safely work in an analytical lab;
2. perform good laboratory practices (GLP);
3. use of Excel for calculations;
4. use of spectrometric techniques (AA, IR, UV)
5. use of chromatographic techniques (GC, HPLC).

Objectives

Thirteen measurable objectives are recognized as essential to the course; laboratory skills are measured from seven lab reports; communication skills are evaluated by the oral presentation; Final comprehensive exam includes questions to measure mastery of each of the ten objectives. A grade of "B" or above is considered mastery.

		Objectives and outcomes
1	Titrimetric calculations	1) Stoichiometric calculations 2) Experimental errors
2	Standard analytical laboratory skills	1) Proper use of analytical balances pipets and burets
3	Write Redox equations	1) Oxidation and reduction 2) Oxidation numbers 3) Balancing redox equations
4	Atomic Absorption Spectrometry	1) Beer's Law 2) Use of Excel 3) Plotting graphs using Excel
5	Ultraviolet spectrometry	1) Electromagnetic radiation 2) Frequency wavelength and velocity 3) Photons 4) Use of proper sample cells 5) Chromophores 6) Chemical structure and light absorption
6	Visible spectrometry	1) Why some chemical compounds are colored? 2) Mixture analysis
7	Fourier Transform-Infrared Spectroscopy	1) Infrared radiation 2) Wavenumbers 3) Molecular vibrations 4) Functional group determination

8	Gas Chromatography	1) Chromatographic process 2) Mobile and stationary phases 3) Injectors and detectors
9	High Pressure Liquid Chromatography	1) Mobile and stationary phases 2) Silica gel 3) Modified silica gel 4) Normal and reversed phase 4) Injectors and detectors
	Spectrofluorometry skills	1) Fluorescence
11	Polarimetry interpretation skills	1) Plane polarized light 2) Specific rotation
12	Laboratory notebook	1) Maintenance of a proper laboratory notebook 2) Good laboratory practice
13	Communication skills	1) Making a professional presentation using PowerPoint

Department of Chemistry, Chemical Biology, and Biomedical Engineering

CH-362
Fall 2022

Instrument Analysis I
Lecture & Laboratory Manual

Lecture:
MON-WED-FRI 09:00 AM – 09:50 AM (Kidde 360)

Laboratory:
Room 429 McLean Building
Section A: MON 2:00 PM – 5:50 PM
Section B: TUE 2:00 PM – 5:50 PM
Section C: Thursday 11:00 PM – 2:50 PM

Professor Athula Attygalle 312 McLean Building 201-216-5575 aattygal@stevens.edu	**Office Hours:** Wednesdays 10:00 AM – 11:00 AM

Teaching Assistants	
Idris Junaid **(isamaras@stevens.edu)**	**Office Hours:** **One hour** before each lab session
Oshane Annon **(oannon@stevens.edu)**	**One hour** before each lab session

TABLE OF CONTENTS

SAFETY RULES

The following regulations are an absolute necessity for a safe laboratory. Please read them carefully, as signing the slip below indicates your agreement to abide by these rules. Since there are personal hazard risks associated with non-compliance (fire, explosions, burns, etc.), you may be asked to leave if you don't follow these rules.

1. Safety goggles must be worn at all times in the laboratory (TAs have been instructed to subtract 10 points for each failure).
2. Report any accidents immediately to your instructor.
3. No eating or drinking in the laboratory.
4. Do not sit on bench tops.
5. Clothing must be appropriate for the lab; no shorts, short skirts, sandals, flip-flops, tank tops, or high heels. Confine long hair and loose clothing. Wear shoes at all times in the laboratory but do not wear: * sandals * perforated shoes. * Always wear long-sleeved and long-legged clothing. While performing laboratory work, never wear short-sleeved T-shirts, short skirts, or shorts. Jewelry that interferes with gloves and other protective clothing, or that could come into contact with electrical sources or react with chemicals should not be worn.
6. Long hair must be tied back. It is advisable to minimize the use of hair sprays and other hair products, because they are highly flammable. Contact lenses should never be worn in the laboratory.
8. Be aware of the location of the safety shower, eyewash station, fire extinguisher, first aid equipment, and the exits.
9. All experiments must be approved by the instructor.
10. Know the hazardous properties of the chemicals you are using. Specific instructions for dealing with hazardous material will be given by the instructor prior to their use. Read MSDS (Material Safety Data Sheets) information. When in doubt, ask! Use gloves appropriately. Note that if you use gloves to handle hazardous chemicals, your gloves are contaminated. **Do not handle clean equipment with contaminated gloves**. **Do not operate computer and instrument keyboards with contaminated gloves. To carry a chemical bottle use only one hand with a glove on. Use the other hand without a glove to do other tasks such as opening doors. Remove gloves when writing notes.**
    - Use the fume hoods when instructed.
    - Dispose of hazardous waste as instructed. No chemical should be poured to municipal drains.
    - Do not pipet by mouth.
11. Use only the chemicals called for in your experiment. Make sure you know what chemicals you are looking for. Many chemicals have similar names or formulas. Treat all chemicals as hazardous.
12. All substances must be properly labeled.
13. No chemicals, supplies, or equipment may be taken from the laboratory.
14. You may not work alone in a lab.
15. Follow your instructor's procedure when pushing glass tubing through corks and rubber stoppers.
16. Wash your hands before leaving the laboratory.

Other Rules

1. Keep the laboratory floor, benches and walls clean.
2. Users of the Electronic Balance. If you find the balance dirty, you can refuse to use it (and go home). On the other hand, if you must use it (because you have to finish an experiment before a deadline), you must clean the balance yourself. The excuse that someone else is responsible for the dirt is *never* acceptable in a scientific laboratory. If you spill chemicals or other material in the balance itself, or on the bench, you must immediately sweep out the contaminant with a brush before proceeding.
3. "Walkmans," music devices, and cellular phones are not allowed. Go to the corridor if you have to use your cell phone.
4. Instrument computers should not be used for any other purpose (**no web-browsing, down-loading. games, or music; no exceptions to this rule**)

SAFETY RULES – HAND-IN

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8. Instrument computers should not be used for any other purpose (**no web-browsing, down-loading. games, or music; no exceptions to this rule**)

Please sign and return this slip to your instructor.

I have read and understand the chemical laboratory safety rules and regulations, and agree to abide by them.

_____ _____

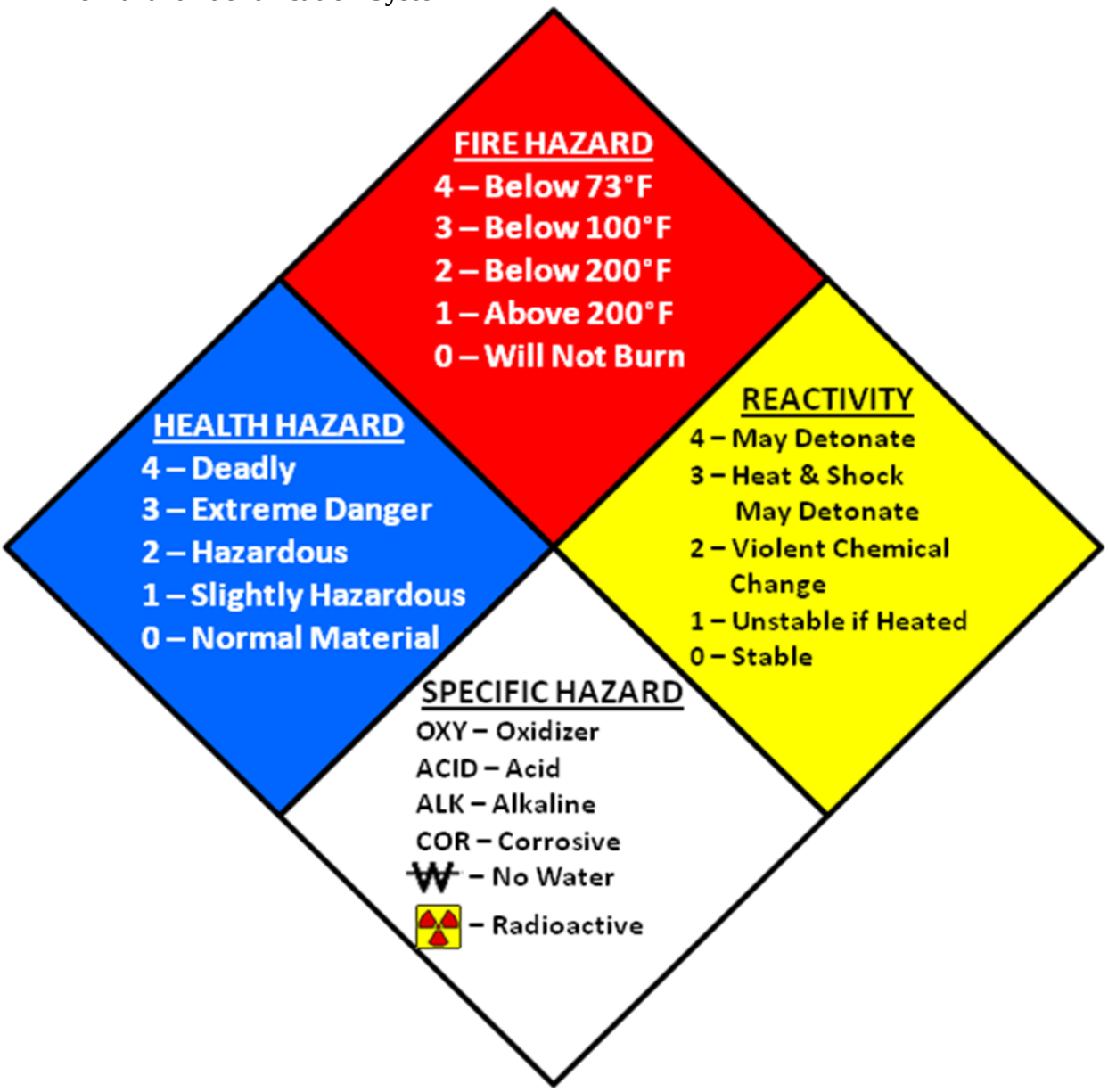
Signature Date CH-362 Fall 2022

MATERIAL SAFETY DATA SHEETS

- You must know where to find MSDS (Material Safety Data Sheets) information for each chemical you use. Visit these links to find the MSDS:
  - Stevens MSDS Website (http://msds.stevens.edu/)
  - Internet MSDS Solutions (http://www.ilpi.com/msds/)

HAZARDOUS MATERIAL CLASSIFICATION

- **NFPA (**National Fire Protection Association) **hazard identification coding system**
- **Fire Hazard Identification System**



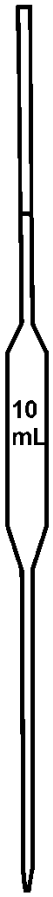
PROPER USE OF AN ELECTRONIC BALANCE

1. Make sure you know what chemical and the amount you want to weigh before going to the balance room. Take your laboratory notebook, and if necessary the appropriate volumetric flask and a wash bottle to the balance room.
2. Find out details about the balance available for your use. You must know the mass arrange and limits of the balance.
3. Ensure the balance is clean, if not clean you can refuse to use the balance, and request the person who used before to clean it.
4. Enter your personal information into the logbook for the balance you will use (this is for Good Laboratory Practice, **GLP**).
5. Tare the balance with the weighing boat. If balance cannot be tared, record the initial weight in your laboratory notebook.
6. Remove the weighing boat from the balance and place it on a clean piece of weighing paper. Add the appropriate amount of the substance to be measured.
7. Check the mass, if an adjustment is necessary, remove the weighing boat, and add or remove substance then measure again. All removal or addition of a chemical should be done outside, not while the weighing boat is on the pan.
8. Record the weight in your lab notebook.
9. Ensure the balance and surrounding area is clean. If you spill a chemical, make sure that you clean it right away. Note in the in the balance logbook that you left the balance area is clean.

PROPER PIPETTING TECHNIQUES

There are different kinds of pipets. In an analytical chemistry laboratory you will be working with fixed-volume, and measuring (delivery) pipettes. Biochemists sometimes use serological (blow-out) pipets to measure small volumes (microliters).

A 10 mL measuring pipet.

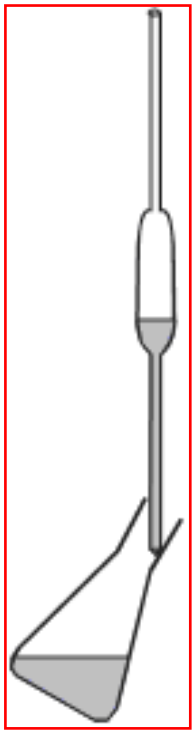


A fixed-volume pipet.

Each pipet needs a different handling procedure. You must know what kind pipet you are using for your experiment so that you do not deliver an incorrect volume of liquid. The measured volume of liquid from fixed-volume pipets is released by gravity, leaving behind a tiny amount of liquid in the tip of the pipet. With **measuring** (delivery) pipettes, the release of liquid must be carefully controlled. After the desired amount of liquid is released, the flow must be stopped at the correct mark. With **serological** (blow-out) pipets on the other hand, the entire liquid volume is delivered by emptying the pipette by force.

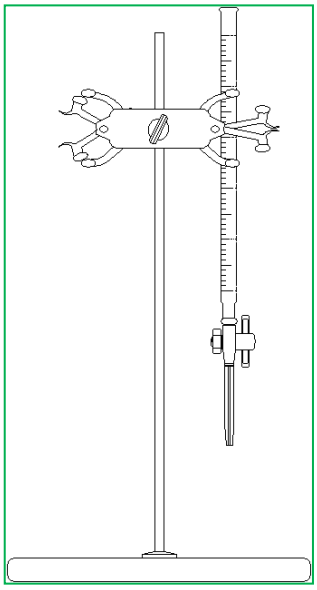
PROCEDURE FOR FIXED VOLUME PIPETS

1. Clean pipet thoroughly and rinse with deionized water.
2. Drain completely, leaving no rinse-water drops inside. If droplets cling to the wall, the pipet cannot be used to execute a quantitative transfer. Clean more thoroughly. If the pipet is wet with rinse water, rinse three times with a few milliliters of the solution to be used in the analysis. DO NOT SUCK SOLUTION BY MOUTH.
3. While filling the pipette, keep the tip of the pipet below the surface of the solution.
4. Using a pipet filler pump, draw the liquid up into the pipet until the fluid level is about one inch above the calibration mark. Take care not to allow the liquid reach the pipet pump (pipet filler pumps are usually color coded; red is for the 25.00 mL pipets).
5. When the liquid is about one inch above the calibration mark, quickly remove the suction device and immediately place the **index finger** (not the thumb) of your dominant hand using same hand to hold pipet, over the exposed end of the pipet, closing the open end, to prevent fluid from flowing. Never hold pipet by the pipet bulb (why?).
6. Remove the pipette tip from the solution, and then wipe any excess fluid from the exterior of the tip with a lint free tissue being careful not to touch the bottom of the tip.
7. Hold the pipet vertically, and let the pipet tip touch the inner wall of the volumetric flask, or any other container holding the solution to be measured. Carefully release pressure on the index finger to allow the meniscus to approach the calibration mark. You must practice this procedure until you are comfortable with this technique.
8. When the liquid meniscus reaches the mark, apply pressure to stop the liquid flow, and drain the drop on the tip by touching it to the wall of the container.
9. Wipe the outside of the pipet with a tissue to remove any droplets adhering to the outside walls. (Do not to touch the bottom of the tip).
10. Transfer the pipet to the receiving container. Hold the pipet vertically and touch the inner wall of the receiving container with the pipette tip (you may tilt the receiving container appropriately) and release pressure on the index finger. Allow the solution to drain completely. Wait 10 seconds (count up to ten) to allow complete dispensing of the sample solution. You must ensure that this is a quantitative transfer. There should not be any droplets clinging to the wall of the pipet after the solution is dispensed. Note that **the liquid is allowed to <u>drain out</u>** and **NOT forced out** Therefore, DO NOT remove by blowing the little amount of liquid that remains at the pipet tip.



PROCEDURE FOR BURETS

1. Clean the buret thoroughly and rinse with deionized water.
2. Drain completely and rinse the buret with a few milliliters of the solution to be measured. Repeat two or three times.
3. Attach buret to a clamp.
4. Fill the buret with the titrant. Use a funnel to avoid spillage.
5. Allow a few minutes for droplets adhering to the inner wall to drain down. Ensure that there are no air bubbles trapped either at the tip or on the sides of the buret (drain the buret rapidly if air bubbles are trapped in the tip beneath the stopcock).
6. Slowly open the stopcock, and carefully release the solution to allow the meniscus to approach the zero mark. When the bottom of the liquid meniscus reaches the zero mark, stop the liquid flow. Wipe the outside of buret tip to ensure no liquid lies on the external surface of the buret.
7. Slowly open the stopcock with one hand and swirl the flask with the other hand or use a magnetic "flea" to stir the solution. As the end point of the titration approaches, decrease the flow rate so that only one drop at a time is delivered.
8. Touch the inner wall of the Erlenmeyer flask to the buret tip to recover a fraction of a drop when necessary.
9. At the end point, close the stopcock and measure the reading from the bottom of the concave meniscus. Make sure that your line of sight is perpendicular to the length of the buret and parallel to the bottom of the meniscus when the reading is taken.



GRADING POLICIES

- CH-362 consists of laboratory sessions and lectures.
- Fifty percent (50%) of your final grading will be based on quizzes and exams from lectures.
- Fifty percent (50%) of your final grading will be based on your laboratory session grades.

GRADING FOR LECTURES

Lectures: Grading Scheme (Total 750 pts).

- Three quizzes. The worst quiz score will be dropped, and the rest will be summed for the final grading. Five random spot quizzes will be given during the class.

Quiz # 1	100 pts
Quiz # 2	100 pts
Quiz # 3	100 pts
Homework (50 x 4)	200 pts
Spot Quizzes (20 x 5)	100 pts
Total (Quizzes)	500 pts
Prelim	100 pts
Final Exam	150 pts
TOTAL	**750 pts**

GRADING FOR LABORATORY

Laboratory: Grading Scheme (Total 750 pts).

Grading (per unit)

	Points
Pre-Lab Report submitted on time. (Due one week before the DAY of the experiment.)	2
Pre-Lab Report contents	8
TA Evaluation	5
Final Lab Report. (Due one week after the experiment)	50
Post-lab questions	15
No Excel sheet	Minus 5
Late Submissions • **Late submissions of Pre-Lab Reports and Final Lab Reports will not be accepted under any circumstances.** • **Missing sections, such as Abstract or Excel Sheets will not be accepted at a later date. The Reports will be graded as received.**	
Total per unit	**80**

Final Grading for Lab Work

Seven Units (80 x 7)	**560**
Lab Notebook. **(Submit all lab notebooks to your TA on or before December 12th, 2022)**	**90**
Oral Presentation	**100**
Total	**750**

Last day for your Final Lab Report is December 12th, 2022

PRELIMINARY LAB REPORTS

The mission of this course is to develop good analytical thinking and problem solving capabilities in students. Students are required to plan their work prior to their arrival in lab. Many calculations that are necessary for the experiment must be done before coming to the lab session. If you have to do these calculations after coming to the lab, you will not have enough time to finish experimental procedures. Each student is required to write a preliminary lab report for each unit. **Each student must write an independent pre-lab report.** A pre-lab report is worth 10 points and is two days before the DAY of the experiment. TAs will grade and return the prelab on the same day it was submitted. TAs will not allow you to proceed with any experiment without a pre-lab report. Two points are awarded if it is turned in on time, zero if it is late. The other 8 points depend on the quality of your report, as discussed below.

The preliminary reports should include the following seven sections. Where appropriate, write complete sentences and use proper grammar. This exact format MUST be followed for full credit.

1. Title of the Experiment. The title should properly describe the experiment.

2. Name of Technique:
    A. Present a schematic (block) diagram of the instrumental technique that shows its essential components.
    B. Write in your own words a brief summary describing how the technique works. If you cut and paste from Web pages give proper acknowledgement and make sure you understand the content. You will be penalized if you cut and paste material that you don't understand.

3. Use of the Technique for your Experiment:
    A. What types of samples can be analyzed by this technique? In other words, what physical or chemical properties must samples have in order to be determined by this technique?
    B. What physical or chemical property of the sample is probed in this measurement?
    C. How can the technique be used for qualitative and/or qualitative measurements?
    D. What are the major advantages and limitations of the technique?
    E. Describe the nature of the signal. (What is actually being detected?)

4. Calculations:
    A. Calculate (tabulate if necessary) the molecular weights needed for your estimations.
    B. Give the amounts that will be weighed (tabulate, if necessary); in what volumes they will be dissolved. Show how dilutions will be made if that is required (tabulate if necessary).

5. References: Include complete citations showing title, author, publisher, year, etc. You must use at least two sources. Follow the format used by the journal "Analytical Chemistry." Here are examples of the reference format:
    1. Koile, R. C.; Johnson, D. C. Anal. Chem. 1979, 51, 741-744.
    2. Willard, H. H.; Merritt, L. L., Jr.; Dean, J. A.; Settle, F. A., Jr. Instrumental Methods of Analysis, 6th ed.; Van Nostrand: New York, 1981; Chapter 2.

    If you cut and paste Figures from the WWW, the URL of the reference must be given. Direct cut and paste of text from the internet will get you a poor grade. Explain in your own words, what is required.

6. MSDS: **Safety Issues and Chemical Hazard Information** must be provided for each chemical you will be using. **Use your own words and don't copy and paste the entire MSDS for the chemical.**

7. PRE-LAB QUESTIONS: Provide answers to any Pre-Lab questions asked in the LAB MANUAL?

POST-LAB QUESTIONS

There will be seven post-lab quizzes. 15 bonus points will be given for each quiz. The time will be announced by the TA.

LABORATORY NOTEBOOK

For good laboratory practice (GLP), it is essential to maintain a proper lab notebook. A laboratory notebook permanently records, what was done in the lab. In a research laboratory, it provides legal proof of what inventions were made and when. Students must maintain a hardcopy laboratory notebook. Data should be entered in such a way that any other analyst is able to follow you techniques and results, or reproduce the results if necessary. Neatness and reliability is an integral part of good laboratory practice. Use a book with permanently bound pages. Spiral or comb bound books are not acceptable in court. Make all entries with ink.

Your laboratory notebook belongs to the laboratory. Entries made in the lab note book are intended to show someone else what you have done in the lab on a particular day. It is the place for your data, not for your personal notes. You may use a separate notebook to take notes for writing your final report. There is also no need for a discussion in your laboratory notebook. Ask the following question to yourself to test whether you are maintaining proper records. Could someone else, with a similar technical background, use your notebook to repeat your work?

1. On the title page of laboratory notebook write your name, address, telephone number, email address, name of the course and instructor's name.
2. The first 3-4 pages should be reserved for the Table of Contents. Page number all the other pages.
3. Data must be recorded IMMEDIATELY in the notebook with a pen (no pencils) while acquiring data (the excuse, I lost all my data due to a computer crash is not acceptable).
4. Even if you do the experiment with a partner, each person should have a separate notebook. Each person should enter data individually.
5. Each experiment should have a title (which indicates the purpose of the experiment), and the start date.
6. Write concisely the procedures you executed (neat and legible handwriting; write in third person passive).
7. If the experiment went wrong, write the reasons, and indicate that the experiment was repeated.
8. Data must never be recorded on loose sheets of paper.
9. If you make an error, neatly draw a single diagonal line through the entry, and use a fresh page. Use of Tipp-Ex, ink bleaches and eradicators, and erasures are not allowed. Don't leave blank areas.
10. Pages must never be removed from the notebook.
11. The notebook should be available for inspection at any time.
12. After recording data and **before** leaving the lab, students must obtain the signature of the TA on the page they have recorded data. No entries should be added to the notebook after obtaining the signature.
13. **Submit all lab notebooks to your TA on or before December 12th, 2022. You receive 90 points for properly maintaining your lab notebook.**

Lab Notebook Check list and Grading Codes

- No table of contents (-5 points)
- Whole book is not page numbered (-5 points)
- Any use of pencil (-10 points)
- Pages torn out of notebook (-5 points)
- No date (-1 point)
- Any scribbling to indicate error other than a single line (-5 point)
- Any use of white-out (-5 points)
- Illegible and scribbled handwriting (-5 points)
- TAs signatures not obtained for each lab (-10 points)

Laboratory Skills, TA Evaluations

Weighing Room (Balance Room):

- There should be no crowding in the room. No more than the number of people using the balances should be in the room (at most 3 people).
- You will a weigh a sample and **quantitatively transfer** the material to a volumetric flask. In order to do so, you must bring all the required materials into the balance room:
    - Material that you are weighing
    - Squeeze bottle containing deionized (D.I.) water
    - All glassware necessary (funnel, volumetric flask, etc.)
    - **Notebook and pen** (-5 points if data are written on a loose sheet of paper)
- You will use weighing boats, not weighing paper. All solid materials will be placed into weighing boats **on the counter, on top of a sheet of paper, not on the balance itself** (-2 points if this rule is not followed).
- Keep the balance room clean **at all times.** You may refuse to use an unclean balance, but you also risk of an incomplete laboratory experiment. If you find the area messy, please clean it up.
- Quantitative transfer should be conducted in the balance room:
    - Using your ring finger and pinky of your non-dominant hand, support the funnel in such a way that you can hold the weighing boat with your thumb and index finger of the same hand. Make sure to hold the funnel above the volumetric flask (or appropriate glassware) without the funnel sitting directly onto the flask.
    - Tilt the weighing boat toward the funnel, allowing the weighed material to be washed down into the funnel. To do so, use the squeeze bottle to squirt water onto the weighing boat, making sure **all** the weighed material goes through the funnel into the flask. Wash the back of the boat also. (Never do a dry transfer! That is just dumping the solid material to the funnel)
    - Rinse the funnel with water into the flask as well as the opening of the flask to ensure that all material has been transferred.
    - Once the transfer is done, you may leave the weighing room and finish the rest at the lab bench.

Other evaluations:

- Do not place feet on the walls in any of the instrument rooms (-5 points).
- No goggles (-10 points)
- No eating and drinking in the laboratory (that includes chewing gum) (-5 points)
- No earphones or ear plugs
- No exposed footwear

LAPTOP COMPUTERS

- Bring your Laptop with Microsoft Excel or another spreadsheet program installed to each lab session. Make sure that the computer is working properly and bring a USB flash drive data back-up. Do not rely on your partner's laptop alone as malfunctions can occur.

FINAL LAB REPORTS

After finishing each Unit, a laboratory report must be submitted within one week after the lab. Reports will not be accepted after two weeks. Each report is worth 50 points.

The final lab reports must be typed using a word processing program. When you work with a partner you will share data, however, **each person must write an independent lab report**. The final report should be your own writing. Laboratory reports are also exercises in learning how to communicate in a formal manner. Utmost attention to detail is required. Imagine you are writing a scientific paper. Follow the style of the journal "Analytical Chemistry," (check http://pubs.acs.org.). Follow guidelines given in ACS Style Guide [A Manual for Authors and Editors, Janet S. Dodd, Editor, third edition; it is good idea to purchase a personal copy; ACS Style Guide, by Anne M. Coghill (Editor), Lorrin R. Garson (Editor)].
http://aerosol.chem.uci.edu/intranet/writing/ACS_style_guide.pdf

TITLE: The title should accurately, clearly, and concisely reflect the emphasis and content of the paper. The title must be brief and grammatically correct. The tile should be complete enough to be understood by itself. Copying the title from the instruction sheet is not acceptable.

Bad Tile: "Gas Chromatography"

Good title: "Quantitative Determination of Caffeine in a VegiCola Sample by Gas Chromatography"

How about this title? "Determination of Low Molecular Weight Silicones in Women After Exposure to Breast Implants by GC/MS" (*Analytical Chemistry*, 2001, ***73***, 606)

AUTHOR NAME(S): *Include names of all members of the group. Use first names, initials, and surnames (e.g., John R. Smith).*

AUTHOR ADDRESS: The affiliation should be the institution where the work was conducted.

ABSTRACT: All reports must be accompanied by an abstract (one paragraph). The abstract should briefly state the problem or purpose of the experiment, indicate the theoretical or experimental plan used, **summarize the principal findings (include numerical values you found)**, and point out major conclusions. Experimental and instrumental details are not required in the abstract. The abstract should tell the reader what interesting things were discovered or

determined rather than reporting what was done. It should not be a teaser to entice the reader to read the Results and Discussion section.

INTRODUCTION: One or two paragraphs. Do not describe the whole technique (this should in the pre-lab report) but describe the important points of the experiment.

EXPERIMENTAL SECTION: Describe the procedures undertaken. Follow the style of the journal "Analytical Chemistry." Remember to use THIRD PERSON PASSIVE to describe the procedures.

RESULTS AND DISCUSSION: Present calculations, figures, tables here. Discuss your results and follow through to draw pertinent conclusions.

FIGURES AND CAPTIONS: Each figure must have a caption that includes the figure number and a brief description, preferably one or two sentences. The caption should immediately follow the figure with the format "**Figure X.** Figure caption." All figures must be mentioned in the text consecutively and numbered with Arabic numerals. The caption should be understandable without reference to the text. Place the keys to symbols used in the figure in the caption, not in the artwork. **All Graphs, Charts, Illustrations, or pictorials of any kind must be labeled as a Figure. The label and captions must be placed on the bottom of the Figure and not the top.**

TABLES: Each table must have a brief (one phrase or sentence) title that describes its contents. The title should follow the format "**Table X.** Table Title." The title should be understandable without reference to the text. Put details in footnotes, not in the title. Define nonstandard abbreviations in footnotes. **The Table label and caption must be at the top of the Table.**

Use tables when the data cannot be presented clearly as narrative, when many precise numbers must be presented, or when more meaningful interrelationships can be conveyed by the tabular format. Tables should supplement, not duplicate, text and figures. Tables should be simple and concise. It is preferable to use the Table Tool in your word-processing package, placing one entry per cell, to generate tables.

REFERENCES: Place references at the end of the report. In any case, place your list of references at the end of the manuscript.

Read recommendations given in *The ACS Style Guide,* 2nd ed., available from Oxford Press. Always use complete sentences, coherent statements, and consistent tense throughout the report. Although most word processing programs will discourage you, use the third person passive tense in your writing. That is the style for scientific reporting.

Do not use first person tenses: "I weighed out 10.0 g of bubliomatric acid."

Do not use second person tense: "You weigh out 10.0 g oxalic acid."

Also do not write instructions in the imperative tense: "Weigh out 10.0 g NaCl."

Instead, use the third person passive tense: "10.00 mL of NaOH solution was measured." ("not were measured")

In regular writing, third person passive is considered weak, since action is performed to an object instead of that object actually doing the action. However, for scientific writing, this tense is perfectly acceptable since it is understood that the lab has already been done (past tense action) and the reader knows who did the lab (your name was on the paper).

Common Mistakes

Acceptable	Not Acceptable	Reason
0.05 g	.5g	(note the zeros and blank spaces)
0.1 M solution	0.1 Molar solution	
50 °C	50°C	space
254 nm	254nm	space
A = 5	A=5	note spacing
25- to 30-mg samples		
Five grams of NaCl was added to the solution.	Five grams of NaCl were added to the solution.	grammar
None of the samples were soluble.	None of the samples was soluble.	grammar

FINAL LAB REPORT GRADING CODES

A) No abstract (-5 points)
B) Wrong numerical results, or no results (-5 points)
C) No final (numerical) results given in the abstract (-2 points)
D) Introduction too short, or insufficient description of the technique (-2 points).
E) Experimental. Instrumental names and sources of chemical are not given (-1 point). Incomplete sentences (-1 point). Sentences not in past passive voice (-1 point).
F) Dilutions are not described in detail (-2 points).
G) Figures cut and pasted from external sources without giving proper credit (-2 points).
H) No Excel sheet (-5 points)
I) No cell and row numbers (-1 point)
J) No cell formulas (-2 points).
K) Wrong cell formulas (-1 point)
L) No Figures and Tables within the Results and Discussion section (-2 points). (Statements such as, "Please see attached sheets" are not acceptable)
M) No axes labels in Figures (-1 point for each occurrence)
N) No Figure or Table caption (-1 point).
O) Improper selection of axes scales (-1 point).
P) No units (-1 point for each occurrence)
Q) Wrong number of significant figures (-1 point for each occurrence).
R) Poor experimental results (-4 points).

ORAL PRESENTATIONS

In addition to your scientific skills, you must also develop your communication skills. Each one of you will face an interview at some point of your life. You must be always prepared to answer questions like "what have you learnt," or "what did you do at Stevens?" So, take this opportunity very seriously and learn how to excel at an interview. You must aim to demonstrate that you are better than your peers. Keep your talk simple. Do not present any material that you do not understand. Think about what questions you might be asked and be prepared to answer them. Practice you talk several times. Do not assume that the audience knows already what you are talking about. Avoid phrases such as "how do I say this," "as you know," "I am sorry," ……

Each student will give a 10-min oral presentation. Topics will be given a week before the presentation. Contact your TA for assistance. You are expected to prepare about 5-10 PowerPoint slides and give them to your TA a day before the presentation. The date for the presentations will be announced in November.

Guidelines:

1) Prepare slides for the audience, not for yourself.
2) Do not read directly from the slides.
3) Do not read from cue cards.
4) Do not present any material that you do not understand.
5) Do not speak to the screen. Speak to the audience. Make eye contact.
6) Do not use unnecessary animations.
7) Do not overcrowd your slides. Write in bullet form not complete sentences.
8) Make sure your slides can be read from a distance.
9) Do not use more than one slide per minute.
10) Check for spelling mistakes.
11) Show passion.
12) Use contrasting colors (not blue and black)

PLAGIARISM AND COPYING WORK

Plagiarism is strictly prohibited in this class. The dictionary defines plagiarism as the act of "...stealing and using the ideas, writings, or inventions of another as one's own" or ".... taking passages, plots, or ideas from another and using them as one's own". Everything that is submitted for grading is subject to the Stevens Honor System. Therefore, it is the student's responsibility to ensure that all submissions abide by the proper citation rules. Any information that is included in the pre-lab report, final lab report, lab notebook, and oral presentation must have the proper citations. Blatant copying and pasting of information will not be tolerated. Also, all works submitted must be completed independently, with the exception of having similar data. If it is determined that the works submitted by two or more students closely resemble each other, the Stevens Honor System will be notifies to take appropriate action.

EXPERIMENTAL UNITS

UNIT 0: VOLUMETRIC REDOX TITRATION

This is a practice unit. You will submit pre-lab and final reports. They will be corrected, graded, and returned to you. However, this is a mock grade. It will not be counted for your final grade. Study your evaluation report and discuss with your TA and the professor about the errors and mistakes you have made. Ask about how to improve your grade

INTRODUCTION

Many redox titrations are based on reactions of iodine. An oxidizing agent is added to excess iodide to produce iodine, which is then titrated with sodium thiosulfate, the reducing agent.

$$I_2\,(aq) + I^- \rightleftharpoons I_3^-$$

Iodine Iodide Triiodide

In this experiment, potassium iodate (KIO_3) is used as a primary standard (what is a primary standard?). From a known amount KIO_3, an equivalent amount of triiodide (iodine dissolves in iodide solutions to form triiodide) can be formed.

$$IO_3^-\,(\text{colorless}) + 6H^+ + 6e \longrightarrow I^-\,(\text{colorless}) + 3H_2O$$ **Equation 1** (reduction)

$$2I^- \rightleftharpoons I_2 + 2e$$ **Equation 2** (oxidation)

$$6I^- \rightleftharpoons 3I_2 + 6e$$ **Equation 3** (multiply **2** by three)

$$IO_3^- + 6H^+ + 6I^- \rightleftharpoons I^- + 3H_2O + 3I_2$$ **Equation 4** (add **1** and **3**)

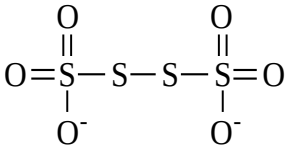
Since I_2 exits as triiodide in the presence of excess iodide, eliminate iodine from **Equation 4**.

$I_2\,(aq) + I^- \rightleftharpoons I_3^-$ **Equation 5** (not a redox reaction)

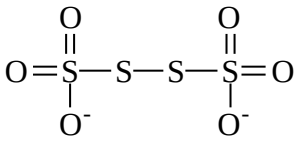
$3I_2 \rightleftharpoons 3I_3^- - 3I^-$ **Equation 6** (multiply **5** by three)

$IO_3^- + 6H^+ + 8I^- \rightleftharpoons 3I_3^- + 3H_2O$ **Equation 7** (add **6** and **4**)

Sodium thiosulfate is a universal titrant for triiodide, which oxidizes it to tetrathionate.

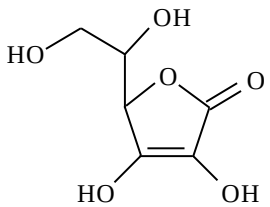
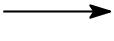
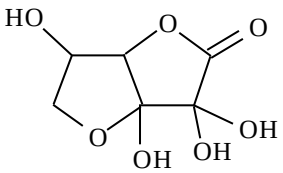
$2S_2O_3^{-2} \rightleftharpoons$  $+ 2e$ **Equation 9** (oxidation)

$I_3^- + 2e \rightleftharpoons 3I^-$ **Equation 10** (reduction)

$I_3^- + 2S_2O_3^{-2} \rightleftharpoons$  $+ 3I^-$ **Equation 11** (add **9** and 1**0**)

As thiosulfate solution is added, the dark brown color of I_3^- solution disappears. Near the end point (pale straw color), a starch solution is added and titrated until the Prussian blue color of the starch-iodide complex disappears. Overall, this procedure is called an iodometric titration (then, what is an iodimetric titration?).

The main objective of this experiment is to use the two standard solutions you have in hand, to estimate the amount of ascorbic acid (vitamin C) present in a commercial tablet. This can be done iodometrically since vitamin C is a mild reducing agent. The idea is to let ascorbic acid from a tablet react with a known excess amount of I_3^-, and then find out the leftover amount of I_3^- by titrating the rest with the standard thiosulfate solution. In this way, the amount of ascorbic acid in a tablet can be computed.

	H_2O			$+ 2H^+ + 2e$	**Equation 12**
$C_6H_8O_6$ 176.13 g/mol Ascorbic Acid			$C_6H_8O_7$ 192.13 g/mol Dehydroascorbic Acid		

PRE-LAB QUESTIONS

1. What is a primary standard? Explain why sodium thiosulfate is not a primary standard.
2. Name three chemicals commonly used as oxidizing agents for redox titrations. Write balanced equations to show how each chemical acts as an oxidizing agent under acidic conditions.
3. Name three chemicals commonly used as reducing agents for **redox titrations**. Write balanced equations to show how each chemical acts as a reducing agent under acidic conditions.
4. What is the difference between iodimetric and iodometric titrations?
5. For acid-base titrations (neutralization), often phenolphthalein and methyl orange are used as indicators for determining the end point. Explain how you would decide the proper indicator for a particular acid-base titration.
6. If two additional drops of water was added when the 50.00-mL solution was made by mistake. What is the v/v percent error introduced by this mistake.
7. A solid mixture weighing 0.05485 g contained only ferrous ammonium sulfate (what is the formula) and ferrous chloride as an equimolar mixture. The sample was dissolved in 1 M H_2SO_4, and the Fe^{2+} present in the solution made in this way required 13.39 mL of 0.01234 M Ce^{4+} for complete oxidation to Fe^{3+}. Write a balanced equation for the redox reaction and calculate the weight percentage of Cl in the original sample.

ferrous ammonium sulfate	$Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$	FM 392.13
ferrous chloride	$FeCl_2 \cdot 6H_2O$	FM 234.84

EQUIPMENT

- Erlenmeyer flasks
- Volumetric flasks (250.00 mL)
- Buret (50 mL)
- Pipet (25.00 mL)
- Pipet filler

REAGENTS

- $Na_2S_2O_3$ solution (supplied by the lab instructor; ~0.06 M; you must determine the exact molarity)
- KIO_3 (solid; read the label on the bottle; note the name of the supplier)
- Starch solution
- Solid KI
- 0.5 M H_2SO_4 solution
- Vitamin C tablet powder

PROCEDURE

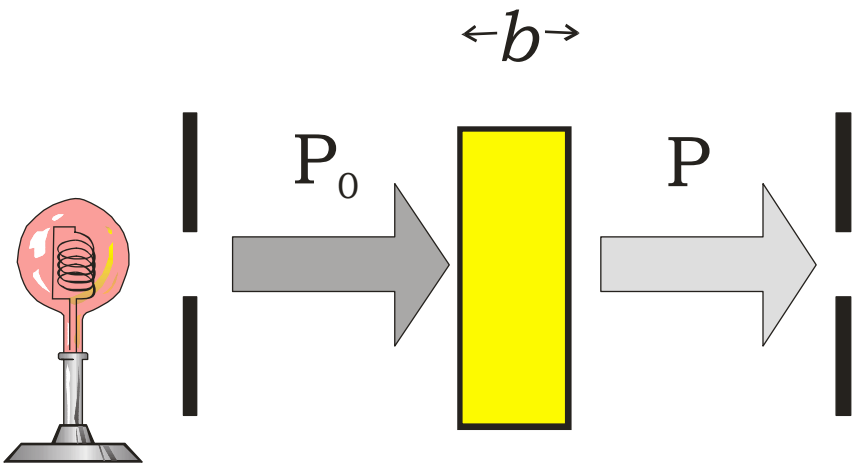
1. Prepare 250.00 mL of **approximately** 0.01 M **standard** KIO_3 solution (you must calculate the amount to weigh and report in the prelab report).
2. Pipet 25.00 mL of the KIO_3 solution you prepared to a 250.00-mL Erlenmeyer flask (this is a quantitative transfer) and add **approximately** 0.5 g of solid KI and **approximately** 10 mL of 0.5 M H_2SO_4.
3. Rinse first, and then fill a 50-mL buret with the $Na_2S_2O_3$ solution provided.
4. Immediately titrate with thiosulfate solution until the solution is pale yellow.
5. Add about 1 mL of starch indicator (why starch?) and titrate carefully until the blue color disappears.
6. Repeat the titration with one additional 25.00-mL portions of KIO_3 solution.
7. Measure approximately 150 mg (0.150 g) of the vitamin C tablet powder.
8. Transfer the powder quantitatively to an Erlenmeyer flask and dissolve the material in 50 mL of 0.5 M H_2SO_4 in (note: some solid biding material may not dissolve).
9. Add 0.5 g of solid KI and 25.00 mL of standard KIO_3 solution. Titrate with standard thiosulfate solution as before. Add starch solution just before the end point.
10. Repeat the ascorbic acid titration one more time.

CALCULATIONS

1. Calculate the molarity of the KIO_3 solution.
2. Calculate the molarity of the $Na_2S_2O_3$solution.
3. Calculate the weight percentage of ascorbic acid present per tablet.

UNIT 1: ULTRAVIOLET-VISIBLE SPECTROSCOPY

INTRODUCTION



$A = \log \frac{P_0}{P} = -\log T = \varepsilon b C$	b = pathlength (centimeters) C = concentration (molar, M) ε = molar absorptivity (M^{-1} cm^{-1}) A = absorbance

Some samples absorb ultraviolet or visible radiation. In absorption spectrometry, we irradiate a sample and measure the reduction of the intensity of the beam due to absorption by the sample. Molecules have preferences to certain wavelengths. Usually, the radiation of this preferred wavelength is selected for quantitative absorption spectrometry. Most light sources produce a whole range of wavelengths. Passing light first through a filter, or a special device called a monochromator, enables us to select the appropriate wavelength required for the experiment.

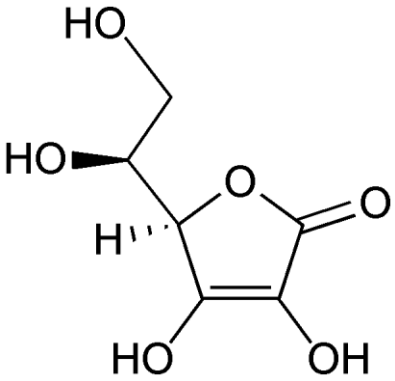
In practice, two measurements of the intensity of light are made. First, the intensity of light of the selected wavelength (λ) reaching the detector when a sample cell (also known as a *cuvet*) is filled with solvent (blank) is measured (P_0). In other words, this measurement represents the concentration of the assayed material when its concentration is zero. Then the absorption by the sample is recorded (P). Molar absorptivity (which used to be called the extinction coefficient) gives us an indication how well a compound absorbs light at a particular wavelength. It is numerically equal to the absorbance recorded by placing a molar solution in a cell of 1.00 cm pathlength.

PRE-LAB QUESTIONS

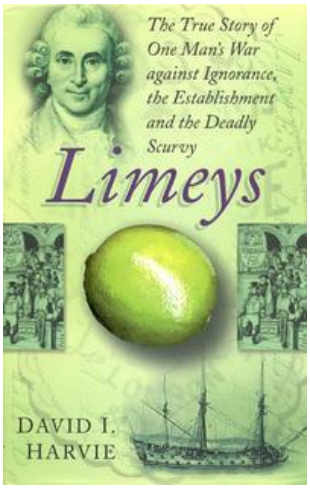
1. What are the units of absorbance and molar absorptivity?
2. Cells made of plastic, glass, and quartz are used for absorbance measurements. Explain how you would select cells appropriate for your experiment.
3. A sample of compound with a molecular mass 292.16 (g/mol) was add to a 5.00-mL volumetric flask. The flask was filled up to mark with water. A 1.00-mL aliquot was withdrawn, placed in a 10.00-mL volumetric flask, and diluted to the mark. The absorbance at 340 nm of this solution was 0.427 when measured in a 1.00 cm cuvet. The molar absorptivity for this compound at 340 nm is $\mathscr{E}_{340}$ = 6130 M^{-1} cm^{-1}.
    a. Calculate the concentration of compound in the cuvet?
    b. What is the concentration of compound in the 5-mL volumetric flask?
    c. Calculate the amount of compound (in milligrams) that was used to make the 5-mL solution?
4. What are UV-A, UV-B, and UV-C radiation?
5. What structural properties are expected to be in the active constituents of a good sunscreen?
6. Explain why some chemicals are colored and some colorless.
7. What is the chemical structure of quartz? Why quartz does not absorb UV light.
8. Laser light is used for some eye surgeries. A commonly used laser for eye surgery emits 544 nm (in air) radiation. The index of refraction of the eye fluid is n = 1.43. Calculate the frequency of this laser wave when it reaches the retina.

Experiment 1.1: Determination of Ascorbic Acid in Vitamin C Tablets

INTRODUCTION



L-Ascorbic Acid
176.13 g/mol



Limeys
The True Story of One Man's War against Ignorance, the Establishment and the Deadly Scurvy

David I. Harvie
2002
ISBN: 0-7509-2772-0

L-Ascorbic acid is an important vitamin. Lack of ascorbic acid in our diet causes scurvy, a disease characterized by weakness, small hemorrhages throughout the body that cause gums and skin to bleed, and loosening of the teeth. Scurvy was a serious problem for English sailors in

the 1600s and 1700s. James Lind, a British doctor, discovered that sailors would not develop scurvy if they were given limes and other citrus fruits. Since this discovery, the Royal Navy made sure that all sailors had lemon juice to drink when they were at sea for longer than one month.

Many believe that ascorbic acid stimulates immune function and protects people from colds and flu-like symptoms. Vitamin C is a water soluble antioxidant, and plays a vital role in protecting the body. Pollutants such as smog and cigarette smoke contain oxidizing molecules that can cause tissue damage. Animals make oxidizing molecules in response to infections. Unfortunately, these molecules while killing infecting organisms can cause tissue damage too. The body requires extra vitamin C when fending off infections. Since ascorbic acid is a water-soluble vitamin, risk of getting an over dose is minimal and any unused vitamin C will be excreted. The minimum daily requirement is 30 mg, and the recommended daily allowance is 60-70 mg per person. For more information, check this website:

http://askabiologist.asu.edu/research/scurvy/

EQUIPMENT

- 25-mL volumetric flasks (5)
- 100-mL volumetric flasks (2)
- 1-mL, 2-mL, 3-mL, and 4-mL volumetric pipets (one of each)
- Matching quartz cuvettes (2)
  - (Why quartz?)
- Perkin Elmer Lambda 25 spectrophotometer

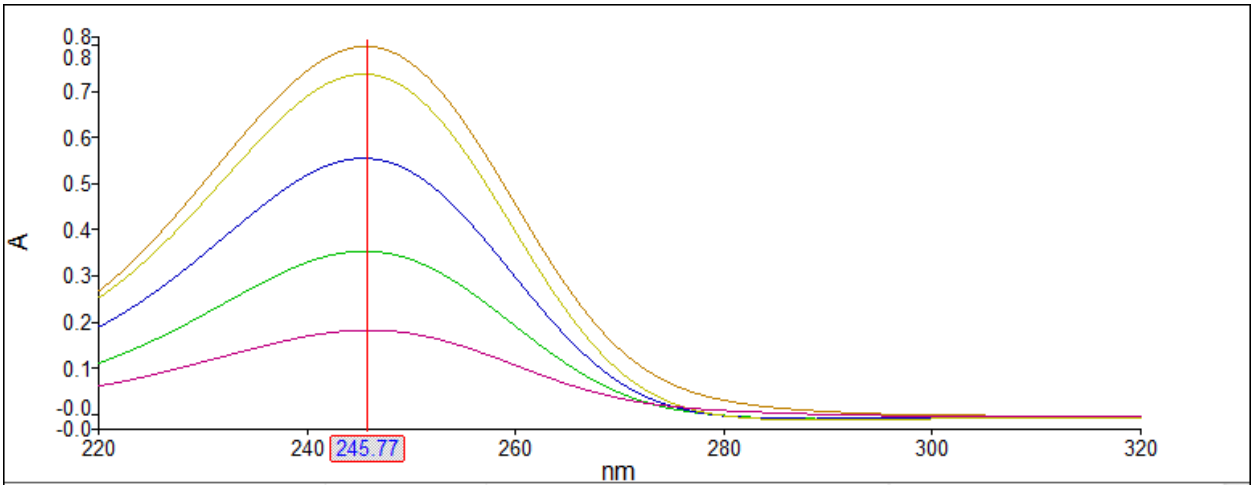
REAGENTS

- L-Ascorbic Acid (vitamin C)
- Vitamin C tablets
- Absolute ethanol

PROCEDURE

1. Clean all glassware before starting the experiment.
2. Prepare a stock solution of ascorbic acid by weighing accurately about 0.010 g (about ten milligrams) of ascorbic acid and transfer it into a 100-mL volumetric flask. Dilute with absolute alcohol to the mark. Stopper the flask and make sure the solutions are well mixed by turning the flask upside down.
3. Pipet 1.00, 2.00, 3.00, 4.00 mL aliquots of ascorbic acid stock solution into four separate 25-mL volumetric flasks and dilute with absolute alcohol to the mark.
4. Fill two quartz cells (**prelab question: why quartz?**) with ethanol. Set the wavelength range of the spectrophotometer from 320 to 220 nm. Open the sample chamber and note that there are two beam positions. Insert one cuvet to the reference slot, and the other to the sample slot and take a blank spectrum (use the auto-zero procedure).

5. Obtain absorption spectra from 320 to 220 nm of all four solutions of ascorbic acid according to the instrument's operating procedure. Make sure to use same cuvet that was used for the auto-zero procedure. Determine the wavelength at which the absorption maximum occurs and record the absorption values for each standard.
6. Weigh accurately a tablet of vitamin C and grind it to a fine powder. Weigh about 0.010 g of this power accurately and transfer it into a 100-mL volumetric flask. Dilute with absolute alcohol to the mark.
7. Pipet a 5.00 mL aliquot of this vitamin C solution, transfer it into a 25-mL volumetric flask and dilute with absolute alcohol to the mark.
8. Obtain its absorption spectrum from 320 to 220 nm of the solution. Record the absorption at the wavelength determined previously. Use ordinary linear calibration method to determine the concentration of ascorbic acid in the unknown solution.



CALCULATIONS

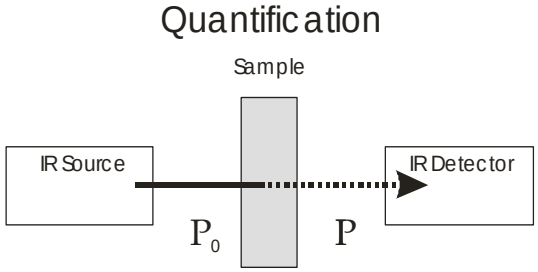
1. Calculate the percentage of vitamin C in the tablet provided.
2. Calculate the molar absorptivity (ε) for ascorbic acid at the wavelength you selected.
3. Discuss what part of the ascorbic acid molecule is accountable for the UV absorption.

1.

UNIT 2: FOURIER TRANSFORM-INFRARED SPECTROSCOPY

THEORY

Nearly all organic or inorganic compounds[1] having covalent bonds absorb various frequencies of infrared radiation. Depending on the type of bonds present, a number of selected frequencies of IR radiation will be absorbed by a molecule.



$$T = \text{Transmittance} = \frac{P}{P_0}$$

$$\%T = \frac{P}{P_0} \times 100$$

$$\text{Absorbance} = A = -\log_{10}\frac{P}{} = \log_{10}\frac{1}{T}$$

With modern FT-IR instruments, it is relatively easy to record spectra from solids, liquids, gases and polymer films. In fact, a choice of sampling techniques exists for all states of matter. Some of the most common sample preparation methods are discussed in this section.

Techniques for Solid Samples

1. A solution in a suitable solvent such as CCl_4.
2. A suspension in liquid (mull).
3. Mixed and pressed into an alkali halide disc.
4. Reflectance techniques.

One of the easiest and the fastest methods to analyze a solid sample is by the mull technique. The solid sample is ground into a fine powder, suspended in a mineral oil such as Nujol, and the resulting mixture is ground to a smooth paste. Then, a small amount of paste is placed in between a pair of NaCl plates and the spectrum is recorded. The disadvantages to this

[1] But not N_2, O_2, or H_2. Why?

technique are the lack of control of the thickness of the paste between the NaCl plates, and scattering of light by the solid particles. The latter can be minimized by using a liquid of the same refractive index as the solid sample.

The traditional technique of analyzing solid samples is by the Pellet Method. In this method, the sample is first ground into a fine powder. The powder is then mixed with dry KBr and ground further. An agate mortar and pestle is normally used to grind the materials (agate is preferred since it is an inert material; KBr corrodes stainless steel). However, using an electrical mill for this purpose is much more convenient. A capsule made of polystyrene, stainless steel, or agate is used to hold the sample during the milling process. Small stainless-steel balls are placed in the capsule with the sample and KBr and a stopper is used to close the capsule. The mill, depending on design, either vibrates the capsule, or rocks it back and forth at very high speed. The finely ground material is then removed from the mill, placed in a KBr die and several tons of pressure is applied with a hydraulic press to coalesce the sample into a transparent or semi-transparent disk. Die sizes range from 1mm, for micro samples, up to the traditional 13 mm diameter disks. When necessary, a vacuum line is connected to the die to remove entrained air, and to a limited extent, entrained moisture. As entrained moisture can cause the finished disk to be opaque the KBr powder should be kept dry by means of a desiccator or a heated oven. Moist samples are not suitable to this technique as they will cloud the disk. Such samples should be dried before analysis.

Assembly and Loading of Sample

Step 1

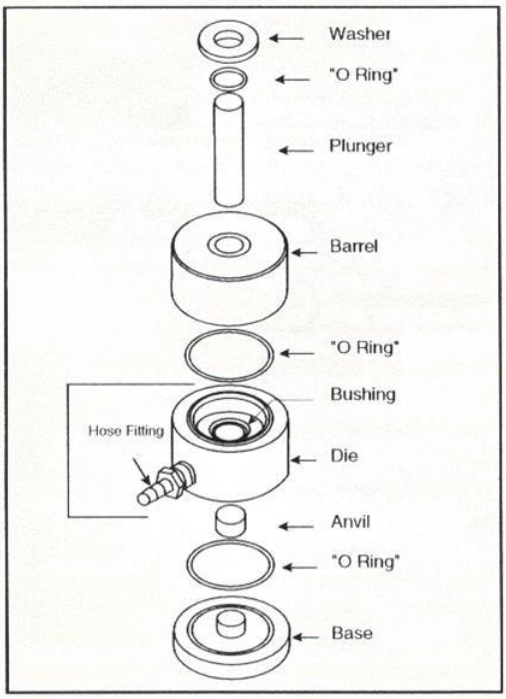
- Place the base on a flat surface and insert the O-ring in the groove.
- Place the die assembly over the post in the base.
- Insert the anvil **POLISHED SIDE UP** into the bushing of the die.

Step 2

- Load the prepared sample through the bushing hole and onto the anvil.
- Level the sample matrix material using a microspatula or a clean, dry, glass rod.

Step 3

- Place the second O-ring in the groove and place the barrel on top.
- Insert the plunger and place the O-ring and washer around the plunger.



Pressing the Pellet

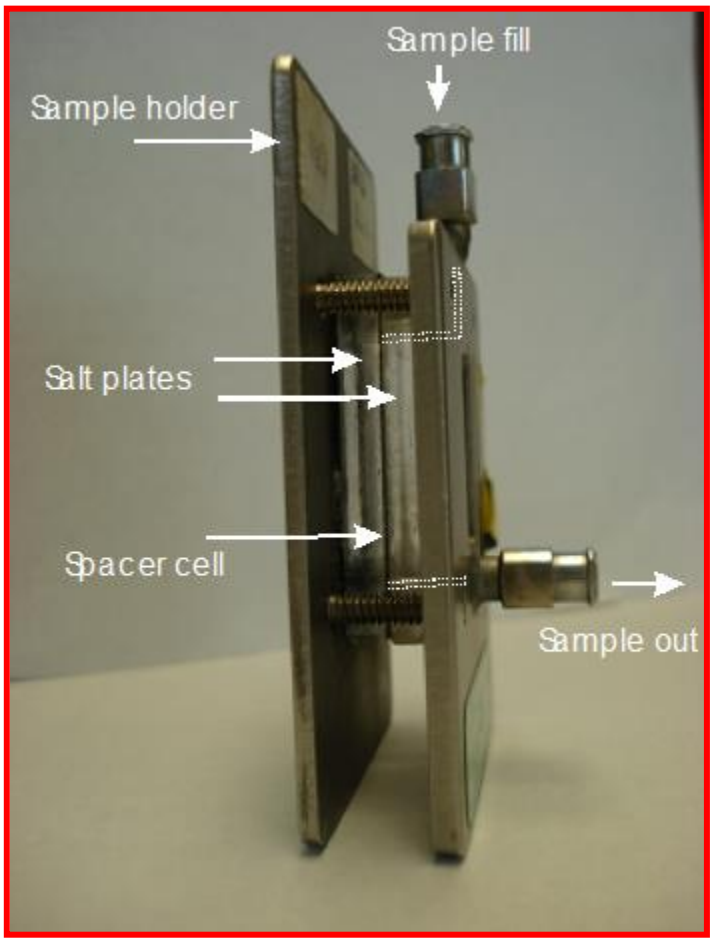
- Place the complete die in a hydraulic press and connect the vacuum line.
- After two minutes under vacuum, press the pellet.
- After an additional two minutes, release the pressure and the vacuum.
- Wait one minute before removing the die from the press.

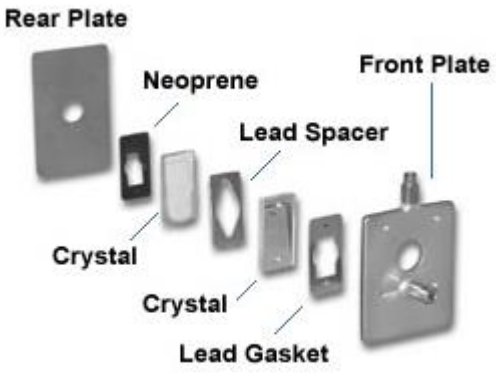
Conversion for 13 mm Die:
lbs. total load = 0.205 x psi
Examples: 25,000 psi - 5125 lbs. total load (gauge reading)
Caution: DO NOT exceed 12,000 pounds total load on the gauge (60,000 psi at sample)

Techniques for Liquid Samples

1. Solution in a suitable solvent such as CCl_4.
2. A thin film of pure liquid between salt plates.
3. Reflectance techniques.

Usually for solutions, a demountable sodium chloride cell is used. The cell consists of two NaCl plates, sandwiching a thin spacer gasket, held together by two supporting metal plates. Two holes drilled into one salt plate, connected to openings in one metal plate allow filling and emptying the cell. The gasket width, which is the nominal pathlength of the cell, may be 1 mm, 0.5 mm, 0.2 mm, 0.1 mm, 0.05 mm, 0.025 mm, or 0.015 mm. Spectra of neat liquids can be recorded as a thin film pressed between two salt plates.

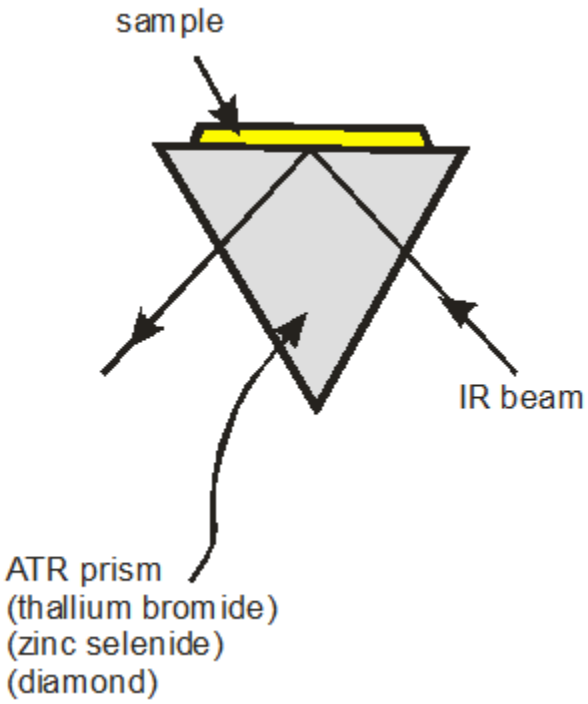




Techniques for Polymer Samples

1. A solution in a suitable solvent such as CCl_4.
2. A thin film.
3. A thin film evaporated on an alkali halide plate.
4. A thin slice cut from larger sample.
5. Reflectance techniques.

Horizontal Attenuated Total Reflectance (HATR) in the Characterization of Polymer Films



In the HATR system, a crystal of an IR transmitting material having a high refractive index, of 2.2 or more, is held in a horizontal plane. When the IR beam from the spectrometer is directed into the crystal at an angle which exceeds the critical angle (which is a function of refractive index), the beam is reflected at the internal surface. However, the beam penetrates slightly beyond the surface before getting reflected. Although, all the energy of the beam is expected to be reflected upon internal reflection, when an IR absorbing sample is placed in optical contact with the reflecting surface the beam loses some energy corresponding to the frequencies absorbed by the sample. In other words, the energy of the beam is attenuated. The beam finally exits the crystal and mirrors of the accessory guide the IR the remaining energy to the instrument's detector. Thus, the attenuated beam when measured can give rise to an infrared absorption spectrum of the material in contact with the ATR prism. For samples of films and polymers, a flat plate system is used. The sample is placed on the crystal and a sample clamp used to provide optical contact. The sample clamp normally has a facility which enables it to accommodate samples of varying thickness and also to provide a range of contact pressures to optimize the experiment, by controlling band intensity.

PRE-LAB QUESTIONS

1. For UV-visible spectroscopy, cells made of plastic, glass, and quartz are used for absorbance measurements. Why such cells are not used for IR spectrometry?
2. What factors determine the magnitude of the characteristic IR frequency of a diatomic molecule?
3. Why water is a bad solvent for IR measurements?

Experiment 2.1: Characterization of Organic Compounds as Thin Films by FT-IR

EQUIPMENT

- Perkin Elmer Spectrum Two

MATERIALS

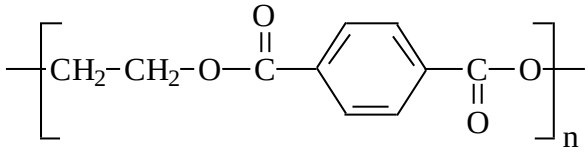
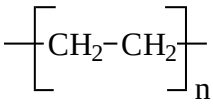
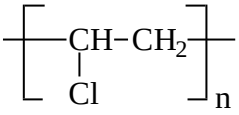
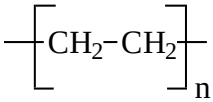
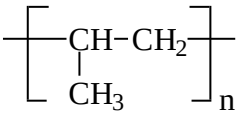
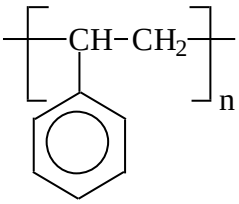
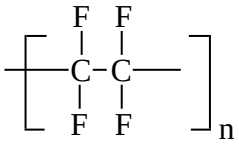
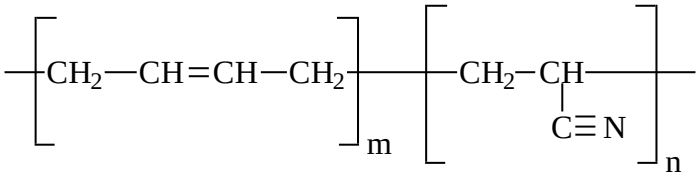
- Polystyrene, polyethylene, nylon, saran, Teflon, polyester, Mylar, and any other available thin films. Soda bottles and cans, and fabrics will also be provided.

PROCEDURE

1. Record infrared spectra of at least five different polymer films, and two kinds of fiber (wool, cotton) by Horizontal Attenuated Total Reflectance (HATR) technique using Perkin Elmer Spectrum Two.
2. Cut one square inch piece from a soda bottle and record its spectrum. Similarly, cut one square inch piece from an aluminum can and record a spectrum from its inner coating.

FINAL REPORT

1. Print spectra and include in your final report. Interpret spectra you recorded. Write the chemical structures of the polymers used. Assign vibrations to least three major peaks in each spectrum.
2. Record spectra from at least three unknown samples. Identify the polymers as best as you can.
3. Write a one-page report on how infrared spectroscopy is used in forensic investigations.

Recycling Symbols					
CODE	**TYPE**	**NAME**	**FORMULA**	**DESCRIPTION**	**SOME EXAMPLES**
PETE	1	polyethylene terephthalate		usually clear or green, sinks in water, rigid, glossy	soda bottles, peanut butter jars, vegetable oil bottles
HDPE	2	high density polyethylene		semi-rigid, sinks in water	milk and water jugs, juice and bleach bottles
PVC	3	polyvinyl chloride		semi-rigid, glossy, sinks in water	detergent / cleanser bottles, pipes
LDPE	4	low density polyethylene		flexible, not crinkly	6-pack rings, bread bags, sandwich bags
PP	5	polypropylene		semi-rigid, low gloss	margarine tubs, straws, screw-on lids
PS	6	polystyrene		often brittle, glossy	Styrofoam, packing peanuts, egg-cartons, foam cups
PTFE					Thread seal teflon
					Nitrile gloves

Experiment 2.2: Characterization of Organic Compounds as Solutions by FT-IR

OBJECTIVE

- The objective of this experiment is to record spectra from neat liquid samples, and then identify functional groups present in an unknown chemical provide by your TA.

EQUIPMENT

- Perkin Elmer Spectrum Two Fourier-Transform Infrared Spectrometer
- Salt plates

REAGENTS

- Cyclohexane
- Ethyl acetate
- Methyl ethyl ketone
- Butanol
- Unknown (labeled UNK? supplied by the laboratory instructor)

PROCEDURE

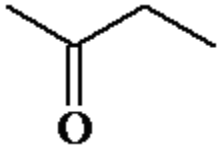
1. Record the infrared spectra of cyclohexane, ethyl acetate, butanol and methyl ethyl ketone.
2. Use salt plates as the background.
3. Add one drop of each liquid onto one slat plate and press with the other salt plate to obtain a thin film.
4. Place the salt plate on the mount and record the spectrum.
5. Carefully wash the plates with $CHCl_3$.
6. Record a new blank spectrum
7. Obtain the infrared spectrum of the unknown compound.
8. Correlate functional groups to absorption peaks observed in all the spectra.
9. Determine the functional groups present in the unknown and identify to which class of compounds it belongs to (e.g. alkanes, alkenes, etc.).

FINAL REPORT

- Present your spectra of cyclohexane, ethyl acetate, and methyl ethyl ketone in an appropriate Figure. Correlate major absorption bands to functional groups of the compounds tested.

Experiment 2.3: Quantitative Determination of Methyl Ethyl Ketone

OBJECTIVE



The objective of this experiment is to determine the concentration of methyl ethyl ketone (MEK) in an unknown sample.

THEORY

In the absence of chemical interactions, infrared absorbencies should obey Beer's Law, $A = \varepsilon \ell c$, where A is the absorbance, ε is the molar absorptivity, ℓ is the path length, and c is the concentration. In this experiment you will prepare a standard curve by measuring the absorbance due to MEK in several standard solutions and plotting the data as absorbance vs. concentration. According to Beer's Law, the plot should be linear. You will then record the spectrum of an unknown sample of MEK and utilize the standard curve to determine the unknown concentration.

EQUIPMENT

- Perkin-Elmer Spectrum Two Fourier-Transform Infrared Spectrometer
- Demountable sodium chloride cell
- Syringe(s)

REAGENTS

- Cyclohexane and methyl ethyl ketone (MEK)
- Solution of methyl ethyl ketone in cyclohexane of unknown concentration (labeled UNK-MEK? supplied by the laboratory instructor)

PROCEDURE

1. Five standard solutions of MEK in cyclohexane of 0.3-1.5% (v/v) range will be provided. Make sure to ask the TA for the actual concentrations. Pure cyclohexane serves as the 0.0% solution. Use the demountable liquid cell having a 0.2 mm gap between the NaC1 windows. The cell is filled using the glass syringe with a steel needle. The cell can be emptied using the plastic syringe to blow out any residual liquid. Clean the cell by flushing it with the organic solvent followed by the solution to be measured. Measure the most dilute solution first. Spent reagents, properly dispose to organic waste.
2. Record a BACKGROUND scan using the empty demountable cell.
3. Record a SCAN for each of the standards and the unknown. Save each spectrum.
4. View the spectra in **absorbance** mode.
5. Identify a peak whose size depends on the concentration of MEK.
6. Overlay the spectra for the six standards and the unknown on one screen. Greatly expand the horizontal and vertical axes to obtain a view showing how the size of the peak of interest varies with concentration. Print this view.
7. Select a frequency that is appropriate for quantification. Measure absorbance values at that frequency from the spectra of standards and that of the unknown.
8. Use Excel to plot a calibration curve and calculate the concentration of MEK in the unknown solution.

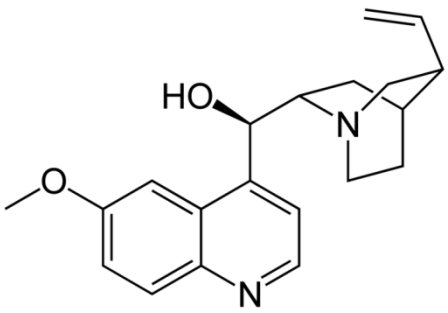
FINAL REPORT

1. Determine the wavenumber of an absorption peak that is suitable for the quantitative analysis? On what basis did you select this peak? What functional group is responsible for this absorption peak?
2. What relationship between absorbance and concentration is revealed from your data?
3. What is the concentration of the unknown?

UNIT 3: SPECTROFLUOROMETRY

INTRODUCTION

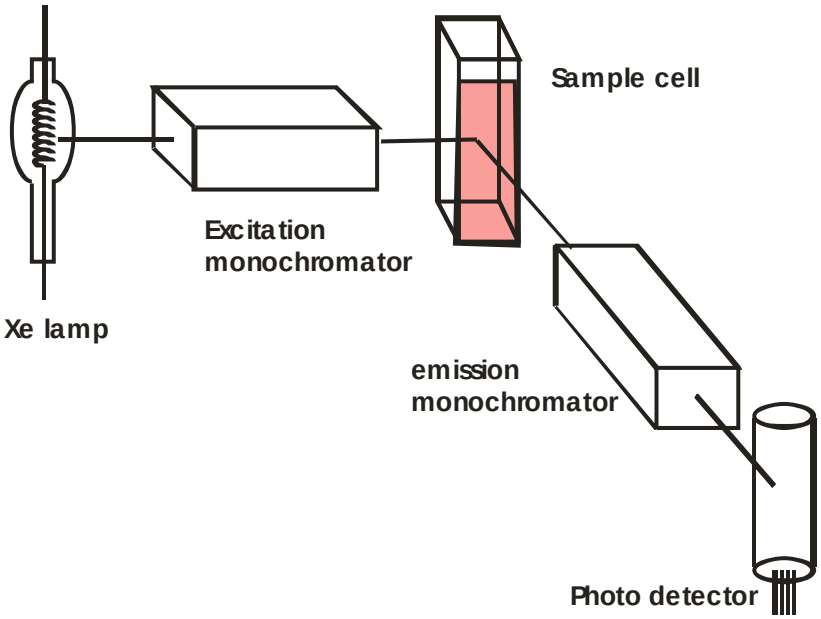
Quinine is an alkaloid extracted from the bark of the cinchona tree, a plant indigenous to the eastern slopes of the Amazonian area of the Andes. Apparently, the name *cinchona* comes from the Countess of Chinchon, the wife of a viceroy of Peru, who was cured of a malarial type of fever by using an extract of the bark of this tree. Although quinine is not used as an anti-malarial drug anymore, it is presently added as the bitter flavoring agent to "tonic water."



$C_{20}H_{24}N_2O_2$ (free base)
Molecular Weight: 324.44 g/mol (free base)
Synonyms: Quinine bisulfate
Quinine hydrogen sulfate dihydrate
CAS No.: 804-63-7
$(C_{20}H_{24}N_2O_2)_2 \bullet H_2SO_4 \bullet 2H_2O$
(quinine hydrogen sulfate dihydrate)
Molecular Weight: 782.96 g/mol
(quinine hydrogen sulfate dihydrate)

Quinine is a strongly fluorescing compound, especially in dilute solutions, and thus can be detected in trace amounts by spectrofluorometry.

A spectrofluorometer is composed of two monochromators. With one the wavelength for excitation is selected. With the other the wavelength of fluorescence is selected. Fluorometric cuvets are similar to those used in regular UV measurements. They are made of quartz. However, fluorometric cells are clear on all sides.

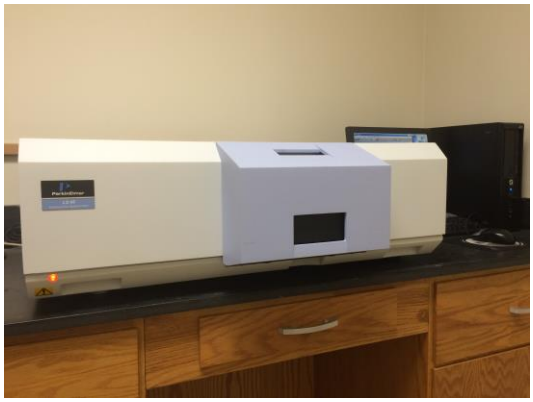


PRE-LAB QUESTIONS

1. Why cuvets used for fluorometric determinations are different from those used for UV measurements?

Experiment 4.1: Determination of Quinine in Tonic Water by Spectrofluorometry

EQUIPMENT



- Perkin Elmer LS-45 Spectrofluorometer
- Fluorometric cuvets (1.000 cm quartz)
- 250-mL volumetric flask (4)
- 100-mL volumetric flask (5)
- Volumetric pipets

REAGENTS

- Sulfuric acid (0.05 M)
- Quinine sulfate
- Commercial tonic water

PROCEDURE

1. Prepare a stock solution of quinine by weighing accurately about 10 mg of quinine sulfate, transferring it into a 250-mL volumetric flask, and filling the flask with 0.05 M H_2SO_4 up to the mark.
2. Prepare five calibration samples of 0.1, 0.2, 0.6, 0.8, 1.0 ppm quinine solution by making appropriate dilutions with 0.05 M H_2SO_4 as the solvent.

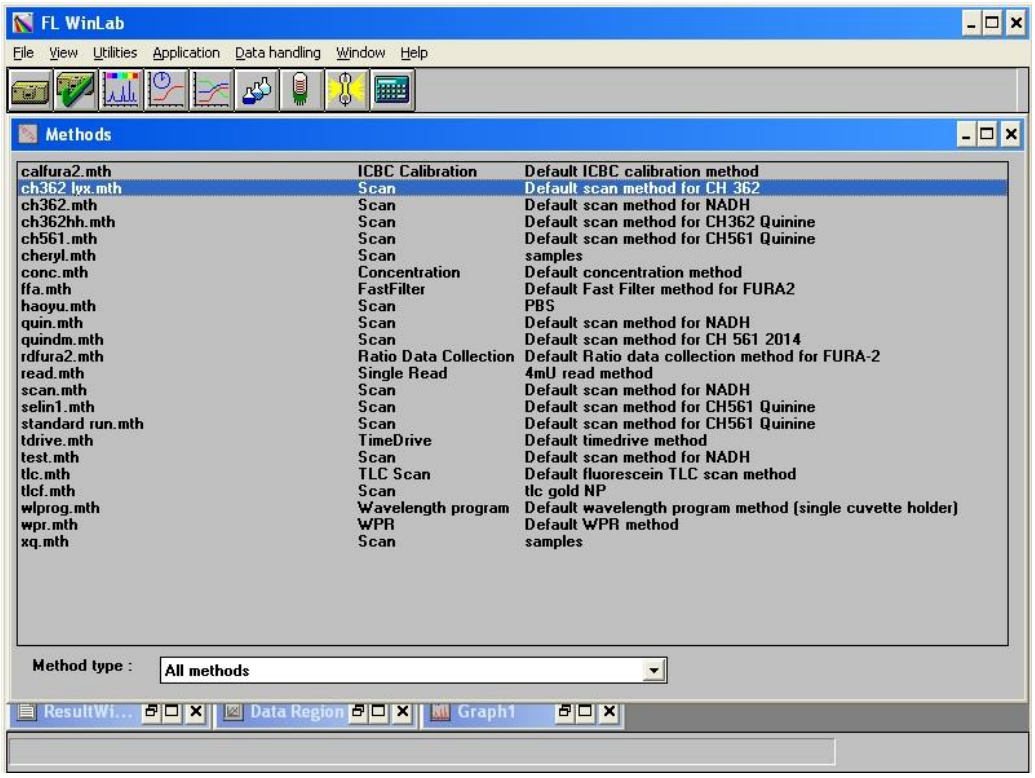
3. Pour about 10 mL of tonic water into a clean and dry beaker and shake it thoroughly to reduce effervescence as much as possible. Then pipette three 1.00 mL samples to three 250-mL volumetric flasks and fill them up to the mark with 0.05 M H_2SO_4.
4. On the spectrofluorometer, set the excitation wavelength to 247 nm and measure fluorescence emission spectrum between 350 and 600 nm of the calibration standard solutions, starting with 0.1 ppm.
5. Measure the fluorescence spectra of the diluted tonic water samples.
6. Construct an ordinary linear calibration curve using the absorbance at the wavelength maxima for each standard. From the absorbance at the wavelength maxima for the tonic water samples, determine the concentration of quinine in tonic water.

CALCULATIONS

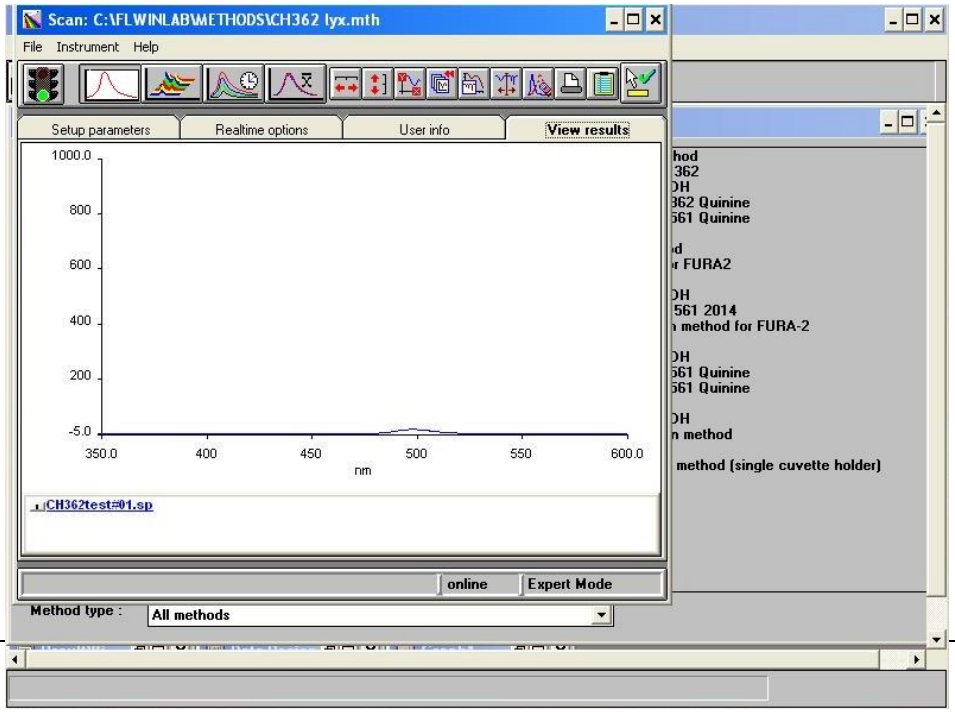
- Construct a calibration curve using Excel. Report the concentration of quinine in tonic water in ppm.

PERKIN ELMER LS 45 SPECTROFLUOROMETER

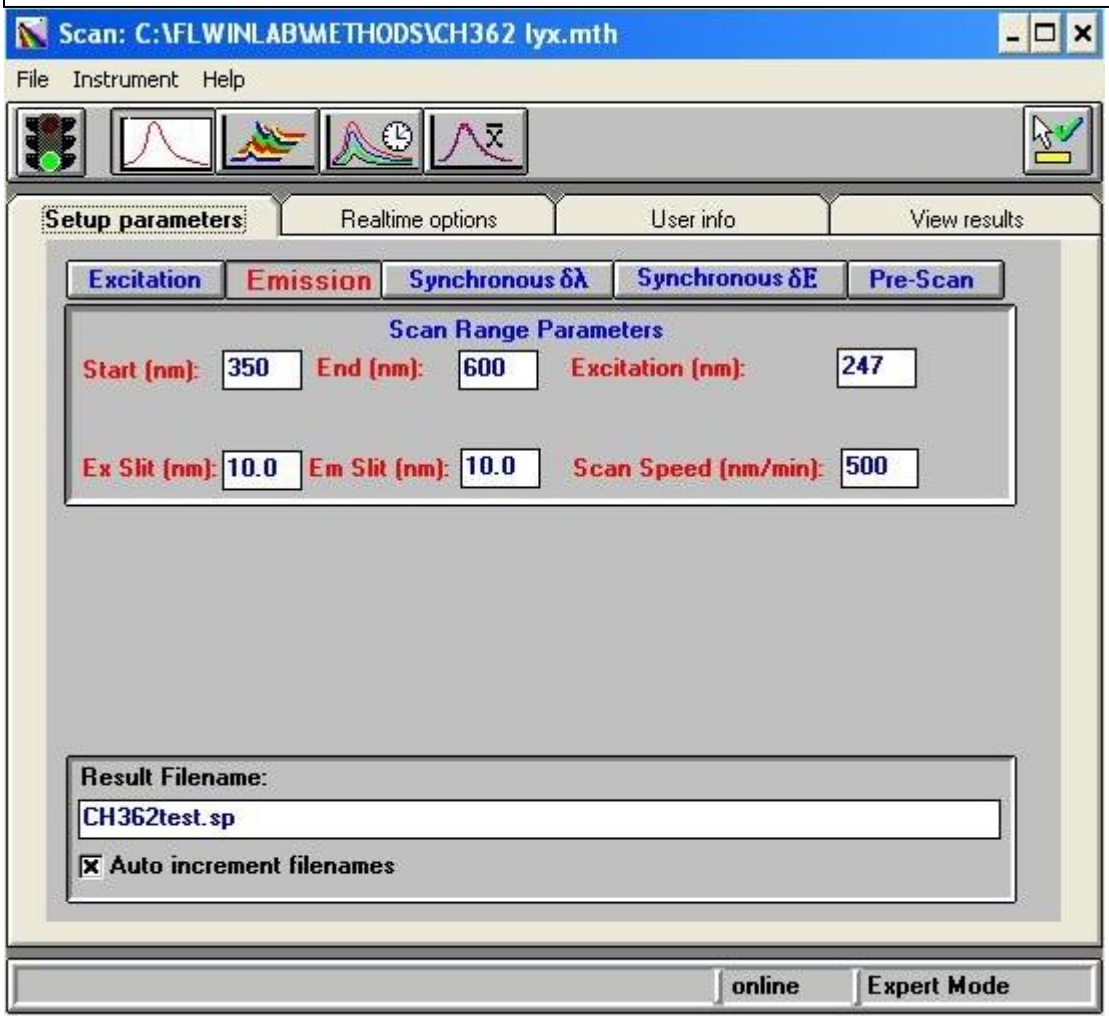
1. Power on
2. Switch on the Xe Lamp (switch is at the left-hand side of the instrument).
3. Start the computer.
4. Open FL WinLabfrom the desktop.
5. Choose “ch362hh.mtd” as the scan method, and double click on that method.



5. The instrument will auto-run a sample, and the spectrum is shown in [**View results**].



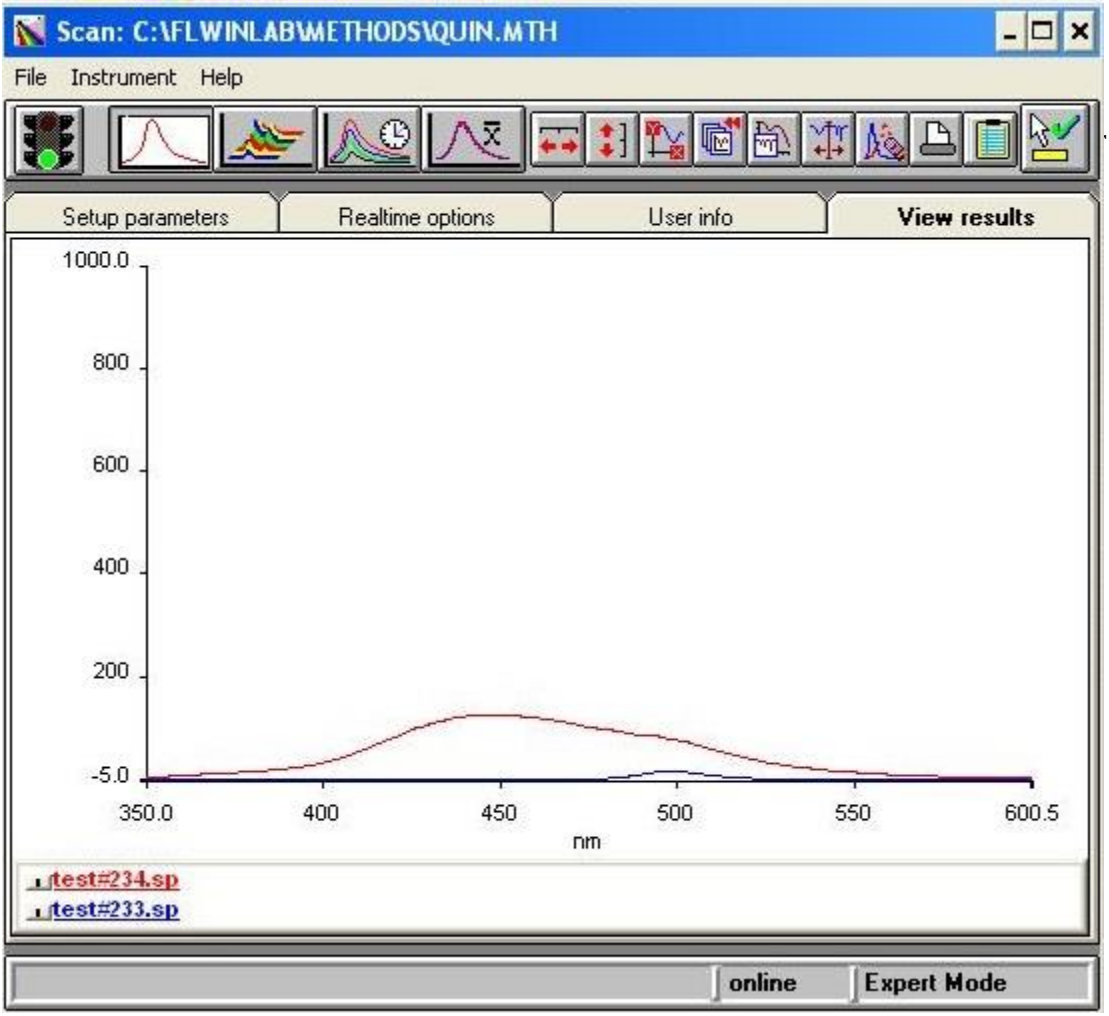
6. Check or modify the parameters for the experiment by clicking on [**Setup parameters**].
7. Parameter setting for the experiment:
   Spectrum type: **Emission**
   Scan Range Parameters:
   Start (nm): **350**; End (nm): **600**; Excitation (nm): **247**
   Ex Slit (nm): **10.0**; Em Slit (nm): **10.0**; Scan Speed (nm/min): **500**



8. Click [**File**] and save the edited method.
9. Fill the cuvet with standard(s) or unknown(s) up to about 80% and insert in the slot of the fluorometer.
10. Background subtraction:
    Fill the cuvet with the solvent and insert in the slot to run a background first.
    Click the **Start** icon at the upper left corner.
    Wait until spectrum scanning is complete, and the spectrum is shown on [**View results**].
    Clicking on [**Realtime options**], check the mark on [**Auto Background Subtract**], and replace the [**Background Spectrum**] file "**subtract.sp**" with the previous solvent sample file name.

11. Recording spectrum:

Fill the cuvet with standard(s) or unknown(s) and insert in the slot.
Click the **Start** icon at the upper left corner.
Wait until spectrum scanning is complete, and the spectrum is shown on [**View results**].
For example, in the [**View results**] window, the blue curve is the background spectrum, and the red curve is the unknown tonic water sample spectrum with auto-subtract of the background.



Attygalle.

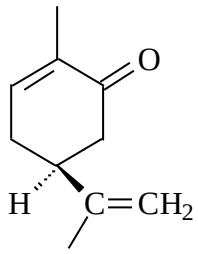
12. Data manipulation:
    Click the **cursor** (the fifth button from the upper right side).
    Move the cursor to the wavelength where the maximum is found.
    Record the wavelength and the absorbance values.

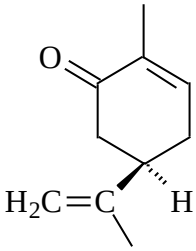
13. Repeat the procedure for all the samples.
14. Ask the TAs to check the data, turn off the software and shut down the spectrometer.

UNIT 4: POLARIMETRY

INTRODUCTION

Foe geometric objects, a structure is considered **chiral** if it is not identical to its mirror image. Some molecules show also this property of chirality. In other words, the overall three-dimensional configuration a chiral molecule cannot be superimposed on its mirror image. Such nonsuperimposable isomers are called **enantiomers.** Such molecules often have a center of chirality. To correctly describe the three-dimensional arrangements of atoms around a chiral center, we use ***R*** and ***S*** nomenclature system proposed by Cahn, Ingold and Prelog. You are expected to know how to assign configurations according to this method using a set priority sequence rules.





S-(+)-carvone bp 230 °C density 0.965 g/mL primary odor component of caraway oil	*R*-(-)-carvone bp 230 °C density 0.965 g/mL primary odorant of spearmint oil

Our understanding of stereochemistry owes much to the initial observations made by French scientist Jean Baptiste Biot in the early nineteenth century. He observed that when linearly polarized light passes through a solution containing chiral molecules, the direction of polarization can be changed. This phenomenon is called **optical rotation** or **optical activity.**

Dextrose is another name for glucose. The name dextrose refers to the fact that it causes linearly polarized light to rotate to the right-hand side (dexter side). Similarly, levulose, commonly known as sucrose, causes the plane polarized light to rotate to the left. Invert sugar, formed by converting sucrose to a mixture of glucose and fructose, gets its name from the fact that the conversion causes the direction of rotation to "invert" from left to right.

In polarimetry, the rotation of polarized light is measured as it passes through a cell containing an optically active solution. The measured rotation can be used to calculate concentrations of solutions. Particularly, substances such as sugars, peptides and some volatile oils can be determined in this way. A polarimeter consists of a polarized light source, an analyzer, a graduated circle to measure the rotation angle, and sample tubes.

The polarized light passes through the sample tube and exhibits angular rotation to the left (-) or right (+). The degree of rotation depends on the wavelength of the light (usually, the yellow sodium D line, near 589 nm wavelength, is used), the optical path length, the concentration of the solution, and the chemistry of the molecule. Under identical conditions, some molecules rotate polarized light more than the others do. In order to measure how good chiral molecules rotate plane-polarized light, we define a term called the “specific rotation.” In fact, the specific rotation is an intrinsic characteristic of a chemical, similar to other properties such melting point, or solubility. By convention, the specific rotation of a chemical is defined as the observed rotation when a plane-polarized light beam of a specified wavelength passes thorough through a 1 g/mL sample in a cell of one decimeter path length (1 dm = 10 cm). Each optically active substance has its own specific rotation as defined in Biots law:

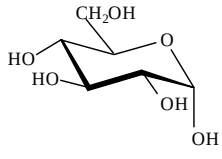
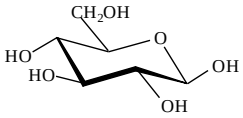
$$a = [a]_D^{20} \bullet \ell \bullet C$$

ℓ = is the length of the cell in decimeters (1 dm = 10 cm)

$[a]_D^{20}$ = specific rotation measured at 20 °C using the yellow sodium D line, near 589 nm wavelength.

C = concentration of the solution in g/mL

Optical properties of some chiral sugars are given below.

Compound	$[a]_D^{20}$
D-glucose (is a mixture of, 36% α-D-glucopyranose and 64% β-D-glucopyranose, which are called anomers)	+52.7º
α-D-glucopyranose  (m. pt. 146 °C)	+112.2º
β-D-glucopyranose  (m. pt. 148-155 °C)	+18.7º
D-galactose (mixture) α-D-galactopyranose β-D-galactopyranose	+80.2º +150.7º +52.8º
Maltose (mixture) α-D-maltopyranose β-D-maltopyranose	+130.0º +173.0º +112.0º
Sucrose	+66.5º

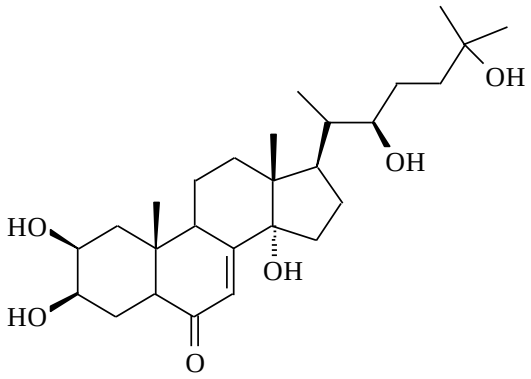
Both anomers of D-glucopyranose can be crystallized and isolated (why are they called anomers?). In fact, they are different compounds with different melting points. However, if a sample of one of these pure anomers is dissolved in water, the optical rotation slowly changes and converges to a constant value of +52.7º. This phenomenon is called mutarotation. Mutarotation of cyclic sugars occurs by a reversible ring opening mechanism. First, the open-chain aldehyde is formed, followed by a ring closure to reform one of the hemiacetal anomers.

For a pure substance in solution, if the wavelength of the radiation and path length are fixed, and the specific rotation is known, the degree of rotation can be used to determine the concentration. The measured rotation can be used for very rapid determination of solution concentrations in a

nondestructive manner. The polarimeter is thus a tool of great importance to those who trade in, or use, sugar syrups in bulk.

PRE-LAB QUESTIONS

1. What is an anomer? How is it different from an enantiomer?
2. Ecdysone is an insect hormone that controls molting. In a polarimetric experiment, 70.00 mg of ecdysone was dissolved in 10.00 mL of chloroform, and the solution was placed in a 2.0 cm path-length cell. The optical rotation was measured at 20 °C using the yellow sodium D line near 589 nm wavelength. If the rotation observed was of +0.087°, calculate the $[a]_D^{20}$ value for ecdysone.



Ecdysone

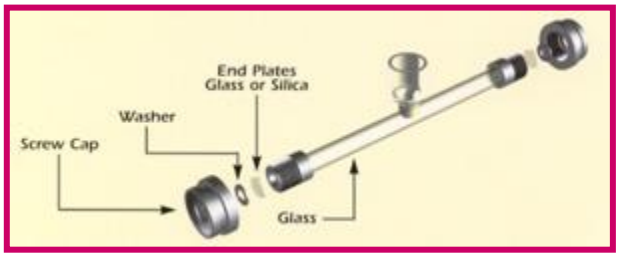
Experiment 4.1: Determination of Sucrose by Polarimetry

EQUIPMENT

- Rudolph Autopole IV Polarimeter (http://www.rudolphresearch.com)
- 250-mL volumetric flask (1)
- 100-mL volumetric flask (5)
- Volumetric pipets

REAGENTS

- Sucrose
- Unknown sucrose solution
- Regular and Diet Beverage



http://www.rudolphresearch.com/cells.htm

PROCEDURE

1. Prepare a stock solution by accurately measuring approximately 25 g of sucrose and quantitatively transferring it to a 250-mL volumetric flask and filling it to the mark with deionized water. This will make a 10% w/v stock solution.
2. Prepare five calibration samples of 1.0, 2.0, 4.0, 6.0, and 8.0% w/v solutions and mix them well.
3. Fill the polarimeter cell with deionized water and zero the instrument. Note the path length of the cell.
4. Fill the polarimeter cell with each solution and measure the rotation using plane-polarized light at 436 nm wavelength.
5. Fill the polarimeter cell with the unknown sugar solution and measure the rotation.
6. Measure optical rotation of the regular and diet beverage (make sure the beverages have been left open for some time to purge all the carbon dioxide out).

CALCULATIONS

- Construct a calibration curve using Excel. Report the concentration of unknown sugar solution in w/v percentage. Find the specific rotation of sucrose. Compare the specific rotation you found with that reported in literature and comment on any dissimilarity you may have observed. Comment on the optical rotation values you recorded with regular and diet beverage.

UNIT 5: ATOMIC ABSORPTION SPECTROMETRY

INTRODUCTION

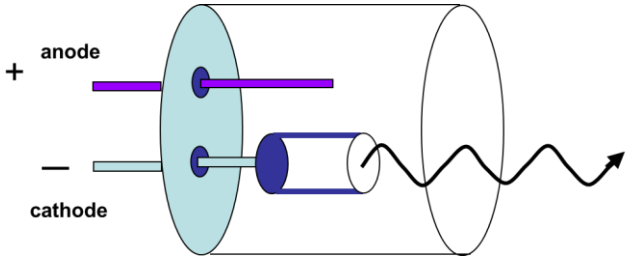
Atomic Absorption Spectrometry (**AAS**) is based upon absorption of radiation by free atoms. Almost any solid, liquid, gaseous sample can be analyzed by **AAS**. However, it is more convenient to convert samples to homogenous solutions prior to use. Usually, a solution is aspirated into a hot flame of about 2000-3000 ^{o}K. In the flame, solvent evaporates and remaining solid is broken into atoms. Usually, a radiation source that emits photons of specific wavelength is selected. The wavelength selected should be specific to the element being determined. When the sample is irradiated, energy is absorbed causing transition of electrons from the ground state to an excited state. By measuring the amount of radiation absorbed, a quantitative estimation of the amount of analyte present can be evaluated.

Basic Instrument Design

The components in an atomic absorption spectrophotometer are:

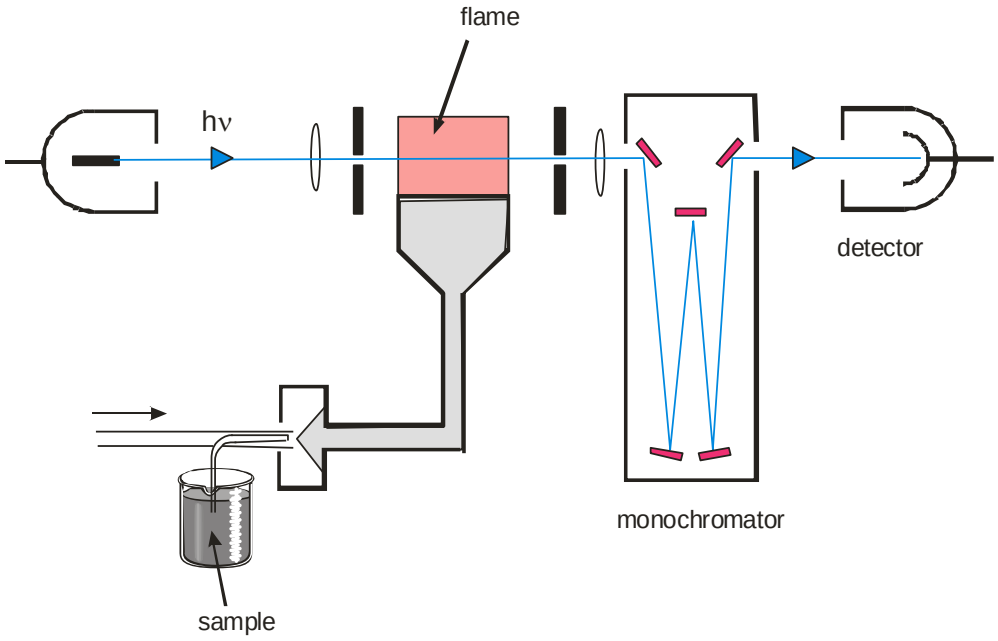
1. A light source (usually a Hollow cathode lamp) emitting monochromatic light specific to the element being determined.

A Hollow cathode lamp



Ca	422.7 nm
Se	196 nm

2. A high temperature flame such as air-acetylene into which a sample solution is aspirated.
3. A monochromator that cuts off radiation all other wavelengths.
4. A photodetector.



PRE-LAB QUESTIONS

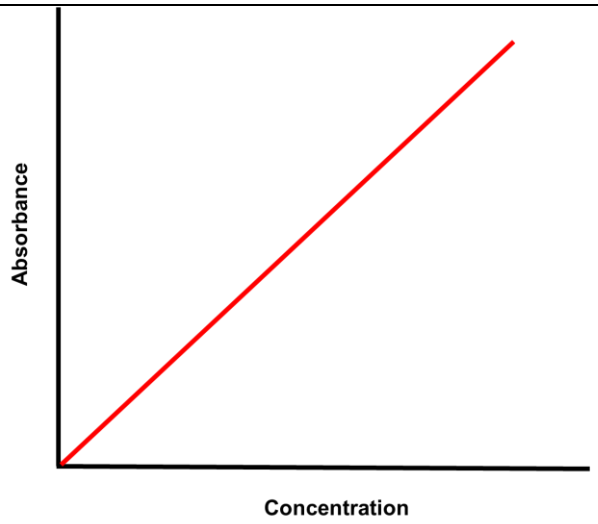
1. How can solid samples be analyzed by Atomic Absorption Spectrometry?
2. What is a monochromator?
3. What is the formula of acetylene? Right three sentences on physical and chemical properties of acetylene.
4. Write a balanced chemical equation to show how acetylene burns in air.
5. For Atomic Absorption Spectrometry, a slot burner is normally used. What is the advantage of using such a long-flame slot burner?
6. What is "hard water"?
7. A series of potassium standards gave the following emission intensities at 404.3 nm. Find the concentration of potassium in the unknown.

Sample (μg K/mL)	Relative emission
Blank	0
5.00	124
10.00	243
20.00	486
30.00	712
UNKNOWN	417

Experiment 1.1: Determination of Copper by Ordinary Linear Calibration Method

THEORY

The concentration of a metal ion in a sample solution can be determined by constructing a calibration plot made from absorbance and concentration data. This technique is generally known as the *Ordinary Linear Calibration Method*. To construct such calibration graph, a series of calibration standard solutions are prepared. These calibration standards should be prepared in the same solvent as that of the unknown sample so that the flow rate of the sample and the standards can be kept identical. A plot of absorbance vs. the concentration of the ion in each solution, according to Beer's law, is a straight line. From this calibration graph, the concentration of the unknown can be found by interpolation.



$$A = \log \frac{P_0}{P} = -\log T = \varepsilon b\, C$$

EQUIPMENT

- Perkin Elmer AAnalyst 200 spectrometer
- 100-mL volumetric flasks (3)
- 50-mL volumetric flasks (4)
- 100-mL beakers (3)
- 1-mL volumetric pipet (1)
- 2-mL volumetric pipet (1)
- 3-mL volumetric pipet (1)
- 4-mL volumetric pipet (1)
- 5-mL volumetric pipet (1)

REAGENTS

- Concentrated HNO_3 (diluted 1.5 HNO_3:1.0 H_2O v/v)
- Concentrated HCl (diluted 1.5 HCl:1.0 H_2O v/v)
- $Cu(NO_3)_2 \cdot 3H_2O$ (primary standard)
- Copper coins (pennies)
- Aqueous sample containing copper of unknown concentration (TAs will give you this)
- Sample of tap water
- Sample of distilled or deionized water (TAs will give you this)
- Sample of drinking water (TAs will give you this)

PROCEDURE

1. Clean all glassware thoroughly before starting the experiment.
2. Accurately weigh and transfer 0.038 g of $Cu(NO_3)_2 \cdot 3H_2O$ (241.60 g/mol) into a 100-mL volumetric flask. Fill the flask with deionized water to prepare a 100 ppm stock solution of copper.
3. Fill a 50.00-mL burette with the stock solution.
4. Prepare six standard calibration solutions (100.00 mL each) by dispensing appropriate amounts of the stock solution from the burette to 100.00-mL volumetric flasks

Sample number	Concentration of copper	Volume of stock solution (mL)	Volume of water (mL)
1	1 ppm		
2	2 ppm		
3	3 ppm		
4	5 ppm		

5. Clean and weigh accurately a copper coin (note the year it was minted). Dissolve the coin completely in *approximately* 10 mL of HCl and 15 mL of HNO_3. **NOTE: These acids are very corrosive. All operations should be carried out in the hood. Gloves and proper safety goggles are mandatory!** Note the color of emanating gases and the bulk solution. When the coin is *completely* dissolved, cool the solution to room temperature and transfer the contents quantitatively to a 100-mL volumetric flask and fill it up to the mark with deionized water. Quantitatively transfer 1.00 mL of this solution to a 100-mL volumetric flask and dilute to the mark with deionized water.
6. The TA will turn on the atomic absorption spectrophotometer according to the instrument's operating manual (observe, and note the roles of different components of the instrument). Write down the instrument parameters in your lab notebooks.

7. The TA will demonstrate how to aspirate your samples. First, adjust the instrument to read zero absorbance while aspirating distilled water. Aspirate your sample solution for 15 seconds and record its absorbance. Once the absorbance is recorded, transfer the solution back into the volumetric flask and pour the next standard solution into the beaker. In this manner, record the absorbencies for all six standard calibration solutions.
8. Construct a calibration curve using the data obtained from the four calibration standards.
9. Aspirate tap water, distilled, deionized water, and the solution given to by your TA and record their absorbances.
10. Transfer the diluted penny solution into a separate 100-mL beaker. Aspirate the sample and record absorbance of this solution.

CALCULATIONS

1. Construct a linear calibration graph by plotting absorbance vs. concentration [μg/mL ≡ mg/L ≡ ppm w/v)] of copper in solution (Use Microsoft Excel; the final Excel sheet should be submitted with your final lab report).
2. Determine the concentration of Cu in tap water, deionized water and unknown sample.
3. Determine the concentration of copper in the diluted penny solution.
4. Calculate the percentage of copper in the coin. Comment and discuss your results. http://www.usmint.gov/about_the_mint/fun_facts/index.cfm?action=fun_facts2

DISCUSSION

- Discuss the chemical reaction between copper and nitric acid. Write a balanced equation. The brown vapor represents what oxide of nitrogen. Discuss the significance of noting the year of manufacture of the coin**. Click here to learn about copper percentage in pennies**.

Experiment 5.2: Determination of Copper by the Standard Addition Method

THEORY

In the S*tandard Addition Method*, known quantities of analyte are added to a sample of unknown concentration. From the increase in signal strength, it is possible to deduce how much analyte was present in the original unknown sample. A calibration graph is constructed and by extrapolating the calibration line, as shown below, to x-axis, the concentration of the unknown can be determined. Mathematically this relationship can be shown as follows:

Let A_0 = Absorbance of unknown when zero amount of known added
A = Total absorbance
a = Absorptivity
b = Path length of light
C_X = Concentration of unknown that we want to determine
C_S = Concentration of standard

Then the following relationship holds true: $A_0 = abC_X$ **Equation 1**

$A = ab(C_X + C_S)$

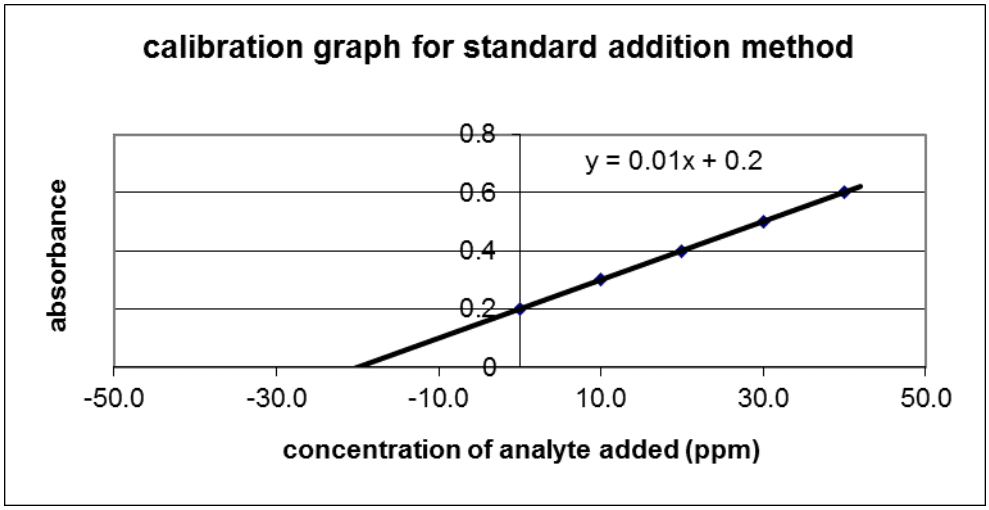
$A = abC_X + abC_S$ **Equation 2**

Substitute equation (1) into (2): $A = A_0 + abC_S$ **Equation 3**

When total absorption (A) is zero then: $0 = A_0 + abC_S$

$-abC_X = abC_S$

$-C_X = C_S$



EQUIPMENT

- 100.00 mL volumetric flasks (4)
- 100 mL beakers (5)
- 1.00 mL volumetric pipet (1)
- 2.00 mL volumetric pipet (1)

REAGENTS

- $Cu(NO_3)_2 \cdot 3H_2O$ stock solution (100 ppm)
- Concentrated penny solution

PROCEDURE

1. Use the concentrated penny solution from the previous experiment. Transfer 1.00 mL of the penny solution into each of the four 100-mL volumetric flasks.
2. Label the first flask as "0.00 ppm" and fill it to the mark with deionized water.
3. Label the second flask as "1.00 ppm" and add 1.00 mL of the 100 ppm Cu stock solution to this flask and fill it to the mark with deionized water.
4. Label the third flask as "2.00 ppm" and add 2.00 mL of the 100 ppm Cu stock solution to this flask and fill it to the mark with deionized water.
5. Label the fourth flask as "3.00 ppm" and add 3.00 mL of the 100 ppm Cu stock solution to this flask and fill it to the mark with deionized water.
6. Measure the absorbencies of these four samples on the atomic absorption spectrophotometer similarly as was done in the previous experiment.
7. Construct a standard addition calibration curve by plotting the absorbance vs. concentration. In this case, the "0.00 ppm" should have a value higher than zero. Extrapolate the line to determine its x-axis intercept, which should give reveal the concentration of the unknown.

CALCULATIONS

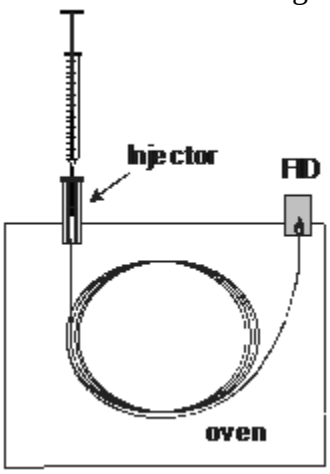
Flask #	vol of 100 ppm Cu solution	total volume	Added Cu conc.		Absorbance
	(mL)	(mL)	ppm	(mL)	
1	0.00	100.00	0.00	1.00	
2	2.00	100.00	2.00	1.00	
3	4.00	100.00	4.00	1.00	
4	6.00	100.00	6.00	1.00	

1. Construct a plot of the absorbencies as a function of the concentration of standard added using Excel. (Be sure to adjust the copper concentration scale to show the intersection of the calibration line at zero absorbance.)
2. Determine the concentration of copper in the concentrated penny solution.
3. Calculate the percentage of copper in the coin. Comment and discuss your results.
4. In your report, compare and contrast the standard addition method to the ordinary linear calibration method. Be sure to comment on the advantages and disadvantages of both methods. Discuss the results of the copper percentage the two methods provided and comment on the method you believe was more accurate.

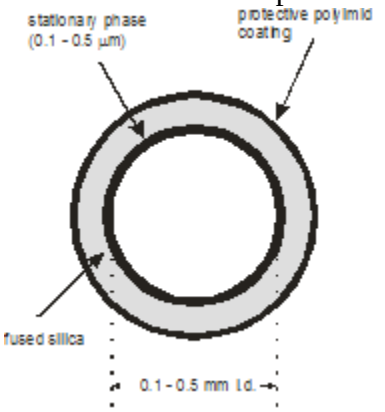
UNIT 6: GAS CHROMATOGRAPHY

INTRODUCTION

Chromatography is a physical method for separating mixtures. Generally, in any chromatographic separation there are two phases (solid, liquid or gas) that move relative to each other while maintaining intimate contact. The moving mobile phase carries analytes with it while the stationary phase tries to retain them. Retention of solutes by the stationary phase will cause solutes to migrate slower than the average velocity of the mobile phase. The extent to which this occurs is reflected in the retention time (R_t, the amount of time required for an analyte to appear at the end of a column of length) of a solute. Since some analytes may stay longer in the stationary phase than others, a separation is afforded.



In gas chromatography, vaporized analytes are transported thorough a column by a gaseous mobile phase (usually helium). The instrument used for the separation is called a gas chromatograph. Basic components of a typical instrument are a heated injector, a precisely temperature-controlled oven, and a detector that responds to minute amounts of analytes that elute from the column. The intensity versus time plot that is recorded is called a chromatogram.



Modern gas chromatography is conducted with open tubular capillary columns made of fused silica. Outside of the columns are coated with polyimide resin to give a certain degree of flexibility to the columns. Inner wall are coated with a very thin film of viscous liquid which acts as the stationary phase.

Useful Websites:
http://www.chem.agilent.com/cag/cabu/whatisgc.htm

PRE-LAB QUESTIONS

1. Clearly define the terms chromatography, chromatograph, chromatogram, and chromatographer.
2. Explain the difference between gas-liquid and gas-solid chromatographic techniques.
3. Compare flame-ionization and thermal-conductivity detection techniques.

Experiment 6.1: Gas-Liquid Chromatographic Separation of a Hydrocarbons

EQUIPMENT

- GC Syringe (10 μL)
- Agilent 6890 Gas Chromatograph equipped with a flame ionization detector.
- HP-5 (5% phenyl methyl silicone) coated fused silica capillary column (30 m x 0.32 mm, 0.25 μm film thickness) (J&W Scientific)

REAGENTS

- A mixture of normal hydrocarbons (*n*-nonane to *n*-hexadecane) in hexane
- Known sample (supplied by the laboratory instructor)
- Unknown sample (supplied by the laboratory instructor)

PROCEDURE

Note column parameters, carrier gas flow rates, and temperature programs used for each experiment.

1. Inject 1 μL of the known hydrocarbon sample into the gas chromatograph while running the temperature gradient program (details will be given by the TA) and record a chromatogram. After finishing the run, integrate the peak areas using the built in ChemStation software program. Record the retention time of the known hydrocarbon.
2. Inject 1 μL of the hydrocarbon mixture sample into the gas chromatograph while running the same temperature gradient program and record a chromatogram. Identify the GC peaks by correlating the retention time of the known hydrocarbon. Inject the mixture two more times.
3. Inject 1 μL of the unknown hydrocarbon sample using the same temperature gradient gas chromatographic conditions and record a chromatogram. Identify the unknown hydrocarbon.

FINAL REPORT

1. Find the average chromatographic peak area for each component (and SD) from the three chromatograms recorded under temperature gradient conditions. Find the mean response per microliter of the solution per component. Plot a histogram with vertical error bars [carbon number x axis, response (= peak areas) y axis]. Comment on the errors observed.
2. Explain the following acronyms used in gas chromatography. WCOT, SCOT, PLOT, FID, and TCD.

Experiment 6.2: Determination of Ethanol in an Alcoholic Beverage

EQUIPMENT

- Gas-tight GC Syringe (10 µL)
- Hewlett-Packard 5890 Series II Gas Chromatograph equipped with flame ionization detector
- PEG column

REAGENTS

- Ethanol
- 1-Propanol
- Unknown sample(s) of alcoholic beverages (beer, whiskey, vodka or gin)

PROCEDURE

Note column parameters, carrier gas flow rates, and temperature programs used for each experiment.

1. Prepare several calibration standards (30% v/v, 40% v/v, 50% v/v, 60% v/v, 70% v/v) of ethanol (in water) in a 50-mL volumetric flask (**show these calculations in the pre-lab**).
2. Accurately transfer 1.00 mL of each solution into separate 3-mL GC vials.
3. Add equal amounts (0.25 mL) of 1-propanol to each of the vials (1-propanol is the internal standard. For more information on the internal standard method, refer to page 49 of the lab manual). **Prelab Questions: what is the concentration of 1-propanol in each of the vials? Also, what is now the concentration ethanol in these vials?**
4. The temperature programs will be given to you during the laboratory session.
5. Inject 5 µL of the headspace for each solution into the GC. Obtain the chromatogram and integrate the peaks using the ChemStation software. Note the integrated values into your notebook.
6. The alcoholic beverage samples will include 1-propanol as the internal standard. Inject 5 µL of each of these samples (probably 3). Obtain the chromatogram and integrate the peaks using the ChemStation software. Note the integrated values into your notebook.

Table 1: Calculation of the Concentration of Ethanol in the Unknown Beverage Sample

Ethanol Conc. (% v/v)	Peak area of ethanol signal (AU)	1-Propanol Conc. (% v/v)	Peak area of 1-Propanol signal (AU)	$\frac{Area_{ethanol}}{Area_{1-propanol}}$	$\frac{Conc._{ethanol}}{Conc._{1-propanol}}$

E_1	A_1	X	B_1	$\frac{A_1}{B_1}$	$\frac{E_1}{X}$
E_2	A_2	X	B_2	$\frac{A_2}{B_2}$	$\frac{E_2}{X}$
E_3	A_3	X	B_3	$\frac{A_3}{B_3}$	$\frac{E_3}{X}$
E_4	A_4	X	B_4	$\frac{A_4}{B_4}$	$\frac{E_4}{X}$
E_5	A_5	X	B_5	$\frac{A_5}{B_5}$	$\frac{E_5}{X}$
E_6	A_6	X	B_6	$\frac{A_6}{B_6}$	$\frac{E_6}{X}$

Read the information on the internal standard method on pages 49 and 50 of the lab manual.

FINAL REPORT

1. Graph the relative concentrations on the x-axis and relative areas on the y-axis. Plot the data points obtained in Table 1. Determine the linear trendline equation. If this line does not intercept the origin (0,0), subtract the y-intercept from all relative areas. Plot these corrected data points and obtain the linear trendline equation. Show this graph in the report.
2. From the trendline obtained previously, the RRF is now known. Using this value and Equation 1 from page 50, the concentration of ethanol in the alcoholic beverages can be determined. Report this value to the correct significant figures.

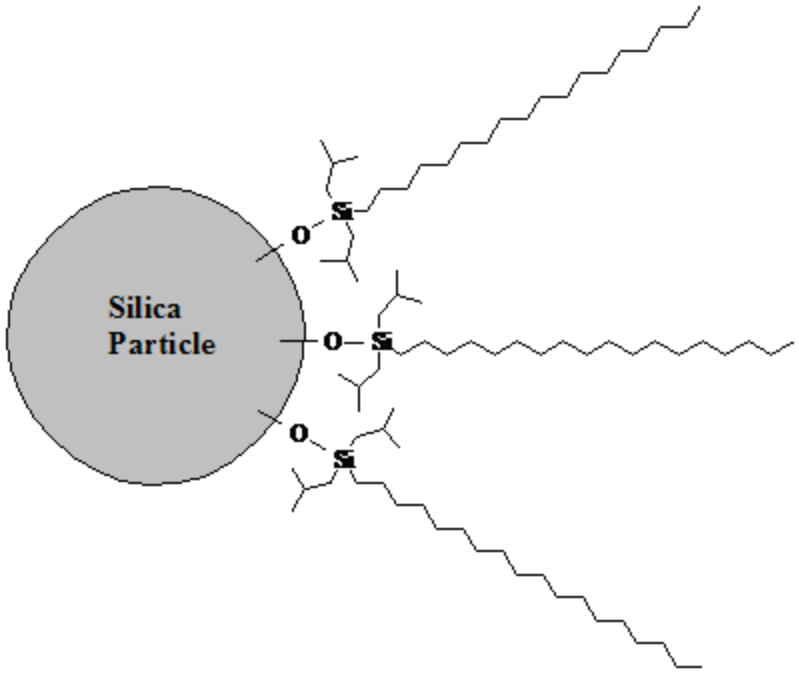
UNIT 7: HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

INTRODUCTION

Modern liquid chromatographic separations are conducted with columns packed with very fine particles (3-5 μm). The use of a high pressure pump is required to force the mobile phase to pass through such columns. This technique is known as high performance liquid chromatography or high pressure liquid chromatography (HPLC).

The two main classes of stationary phase are commonly used in HPLC.

1. **Normal Phase** columns are usually packed with silica gel; they work in the adsorption mode, in a manner similar to that of a silica gel column in conventional gravity chromatography. Since the stationary phase is polar, *a more polar solvent has a higher eluent strength*.
2. **Reversed Phase** chromatography is the most common form of HPLC. Frequently, reversed phase columns are packed with a chemically bonded octadecylsilyl coated silica; such columns are referred to as C-18 RP columns. Thus, the stationary phase is nonpolar or weakly polar and the solvent used is more polar. In this case, *a less polar solvent has a higher eluent strength*.



The following parameters should be considered when selecting an HPLC column.

Length	
Internal Diameter (I.D.)	
Phase	
Particle size	
Pore size	

PRE-LAB QUESTIONS

1. To inject an aliquot of a solution to an HPLC, often a sample loop is used. What are the reasons for employing this technique?
2. An HPLC analysis of a mixture was monitored at 240 nm. The chromatogram obtained showed three peaks. The ratios obtained by peak integration procedure were 1:2:5. Comment of the composition the sample.

Experiment 7.1: RP-HPLC Separation of a Mixture of Compounds

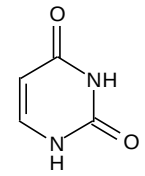
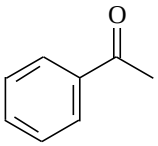
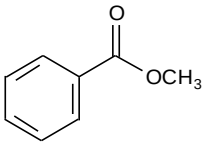
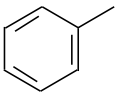
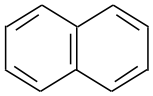
EQUIPMENT

- HPLC Syringe (50 µL)
- Agilent 1100 Liquid Chromatograph with a diode array UV detector

REAGENTS

- Reverse Phase Test Mixture (uracil, acetophenone, methyl benzoate, toluene, naphthalene)
- Filtered HPLC grade solvents (methanol, water, acetonitrile)

Reverse Phase Test Mixture		
Name	**Structure**	**λ_{max} (nm)**

Uracil		
Acetophenone		
Methyl benzoate		
Toluene		
Naphthalene		

PROCEDURE

Note the column parameters (manufacturer, packing material, length, and I.D.), solvent flow rate and pressure, dimensions of the injector loop, and ambient temperature of the lab.

1. Set up an acquisition data file in the computer. Set pump parameters for a 50:50 water:acetonitrile isocratic run. Learn from the instructor how the HPLC injection valve works.
2. **LOAD** 10 μL of the Reverse Phase Test Mixture to the injection loop using a syringe.
3. **INJECT** the sample. Record a chromatogram monitoring UV absorption at 254 nm.
4. View UV spectra of all components. Repeat the injections twice more.
5. Integrate the chromatographic peak areas using the built in ChemStation software program.
6. Repeat the experiment thrice, each time injecting 40 μL of Reverse Phase Test Mixture. Integrate the chromatographic peak areas using the built in ChemStation software program.

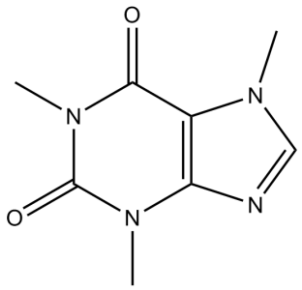
FINAL REPORT

1. Using UV spectra you recorded, assign the chromatographic peaks to respective compounds. Comment on the elution order of compounds. Comment on why absorption maxima [λ_{max} (nm)] are different for each compound (relate to structure).
2. Plot a histogram showing average peak areas and standard errors from 10 μL injections (one set of columns), and average peak areas and standard errors from 40 μL injections (another set of columns).
3. Compare the two sets of columns and comment on the important points (particularly, the standard errors) you observe.

Experiment 7.2: Determination of Caffeine in a Commercial Beverage

INTRODUCTION

About 90% of adult Americans consume caffeine in one form or another every day. In fact, more than half of all American adults consume more than 300 mg of caffeine every day, making it America's most popular drug by far. Caffeine is central nervous system stimulant. Caffeine containing products include coffee, tea, cola, chocolate, and many over the counter analgesics.



$C_8H_{10}N_4O_2$
Molecular Weight = 194.19 g/mol

REAGENTS

- Benzophenone (1.000 mM in water:acetonitrile 50:50)
- Benzoic acid (1.000 mM in water)
- Caffeine (1.000 mM in water)

PROCEDURE

Determining Detector Response Ratios

1. Pipette 5.00 mL of each of 1.000 mM aqueous benzophenone (the internal standard (IS)), 1.000 mM caffeine, and 1.000 mM benzoic acid into a 25-mL conical flask. Mix well. Inject 30 μL into the HPLC injection loop. The TA will help you identify the peaks relative to each component.
2. From the chromatographic trace of the mixture, determine the ratio of the areas relative to that of the internal standard, and hence determine the relative molar response factors for caffeine and benzoic acid relative to the internal standard.

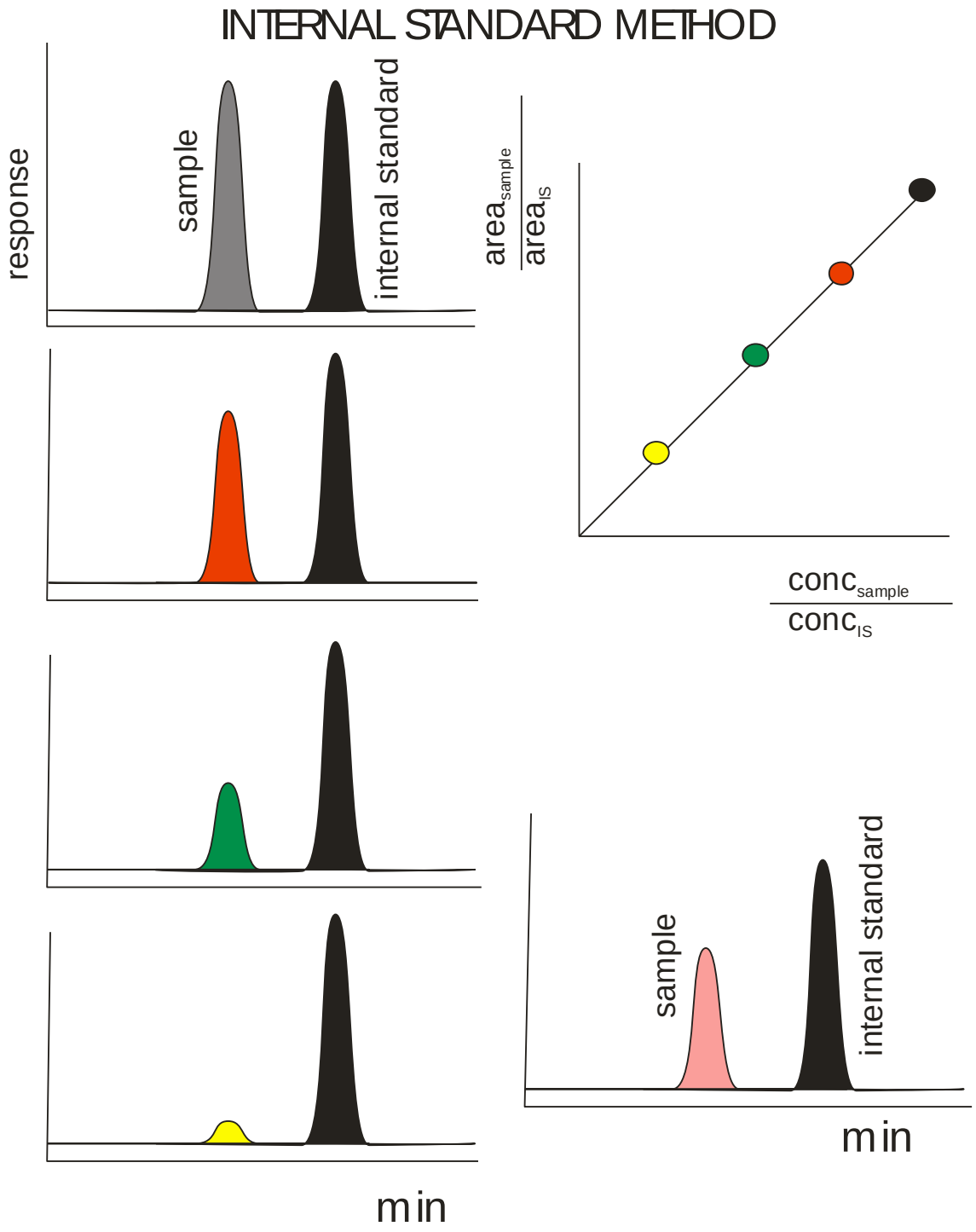
	Peak Area	**Relative Area (Area of Analyte/ Area of IS)**	**Ratio of Conc. (Conc. Of Analyte/ Conc. of IS)**	**Molar Response Ratio**
Caffeine				
Benzoic Acid				
Benzophenone (IS)		1.000	1.000	1.000

Determining the Calibration Curve

1. Prepare four diluted standard solutions of 0.2 mM, 0.4 mM, 0.6 mM, and 0.8 mM of caffeine by pipetting the proper volumes of 1.000 mM caffeine into four 25-mL volumetric flasks.
2. Prepare four solutions by mixing 10.00 mL each of 0.2 mM, 0.4 mM, 0.6 mM, and 0.8 mM caffeine solutions and 10.00 mL of 0.2 mM benzophenone in four separate 25-mL conical flasks.
3. Inject 30 μL of each calibration mixture into the HPLC injection loop. Record chromatograms and determine peak areas.
4. Obtain about an appropriate commercial beverage. Pipet 10.00 mL of the beverage into a 25-mL conical flask and add 10.00 mL of 0.2 mM benzophenone (IS).
5. Inject 30 μL of the above solution and record an HPLC trace. Determine the relative peak areas using the known concentration of the IS.

FINAL REPORT

1. Present data in the form of a Table which clearly shows how the values were obtained.
2. Determine the relative peak area and correct for the molar detection responses of the individual components.
3. Determine the concentration of caffeine in the beverage sample.


INTERNAL STANDARD METHOD
response
sample
internal standard
min
area sample / area IS
conc sample / conc IS
sample
internal standard
min

Since the internal standard and the analyte do not absorb UV light in an identical manner, we must calculate the relative response factor (RRF) as shown in Equations 1 and 2.

Caffeine Conc. (mM)	Peak Area of Caffeine Signal (AU)	Benzophenone Conc. (mM)	Peak Area of Benzophenone Signal (AU)	$\frac{Area_{Caffeine}}{Area_{Benzophenone}}$	$\frac{Conc._{Caffeine}}{Conc._{Benzophenone}}$
E_1	A_1	X	B_1	$\frac{A_1}{B_1}$	$\frac{E_1}{X}$
E_2	A_2	X	B_2	$\frac{A_2}{B_2}$	$\frac{E_2}{X}$
E_3	A_3	X	B_3	$\frac{A_3}{B_3}$	$\frac{E_3}{X}$
E_4	A_4	X	B_4	$\frac{A_4}{B_4}$	$\frac{E_4}{X}$

$$\frac{\text{Area of analyte signal}}{\text{Concentration of analyte}} = RRF \frac{\text{Area of standard signal}}{\text{Concentration of standard}} \quad \textbf{Equation 1}$$

$$\frac{\text{Area of analyte signal}}{\text{Area of standard signal}} = RRF \frac{\text{Concentration of analyte}}{\text{Concentration of standard}} \quad \textbf{Equation 2}$$

The RRF can be determined by a plotting a graph $\frac{Area_{Caffeine}}{Area_{Benzophenone}}$ versus $\frac{Conc._{Caffeine}}{Conc._{Benzophenone}}$. The slope of this graph gives the Relative Response Factor (***RRF***). When the analyte and internal standard are of equal concentrations, the area under the analyte peak is reported as ***F*** times smaller or larger than that of the internal standard peak.

NOTE: Remember that the diluted standards of caffeine and benzophenone were mixed and therefore, the concentrations of these components in the final mixtures are changed. When calculating the RRF, it is important that these correct concentrations are used. This also holds true for the commercial beverage mixture.

In this way, the concentration of caffeine solution that was injected to the HPLC can be calculated. However, what is required is that we know the caffeine concentration in the beverage itself.

APPENDIX

Frequently, the word analysis is used improperly even by experienced chemists. Only samples are **analyzed**. Elements, ions, and compounds are identified in a sample. It is correct to say that an analyst found traces of asbestos in a soil sample from a construction site. It is incorrect to say that asbestos was analyzed unless it means that the sample was asbestos (a so-called "pure" sample) and it was analyzed to determine the presence of impurities.

Unit Prefixes in Metric System

Unit Prefixes in Metric System

Prefix	Symbol	Power of ten
yotta	Y	10^{24}
zetta	Z	10^{21}
exa	E	10^{18}
peta	P	10^{15}
tera	T	10^{12}
giga	G	10^{9}
mega	M	10^{6}
kilo	k	10^{3}
hecto	h	10^{2}
deka	da	10^{1}
		10^{0}
deci	d	10^{-1}
centi	c	10^{-2}
milli	m	10^{-3}
micro	μ	10^{-6}
nano	n	10^{-9}
pico	p	10^{-12}
femto	f	10^{-15}
atto	a	10^{-18}
zepto	z	10^{-21}
yocto	y	10^{-24}

RULES FOR SIGNIFICANT FIGURES

- **Initial zeros** are not significant. Because they are only used to locate a decimal place. The number 0.0777 has three, and 100.001 six significant figures, respectively.

- **Final Zeros**. Zeros after a nonzero digit are significant only if a decimal point is present. Examples. .

Number			Number of Significant Figures
7.77×10^4	=77700	7.77E+04	3
7.700×10^1	=77.00	7.700E+01	4
7.7000×10^2	=770.00	7.7000E+02	5
9.25×10^4	= 92500	9.25E+04	3
9.250×10^4	= 92500	9.250E+04	4
6.532×10^{-4}	= 0.0006532	6.532E-04	4

- **Addition and subtraction**. First carry out addition and subtraction, then round the answer to reflect the appropriate number of significant figures.

Examples:

24.35 + 3.147 27.497 = 27.50	24.3 + 3.137 27.437 = 27.4

Do the following.

65.01 + 0.0000015 =
55.05 - 10 =

- **In multiplying and dividing** two measures numbers, the final product or quotient should bear the same number of significant digits as the least significant quantity in the problem.

Examples:

5.4 X 2.621 = 14.1534 = 14.1
4.234/2.34 = 1.8094017 = 1.81

A sheet of carpet is 2.47 m long and 1.03 m wide. What is its area?

- **Counting and measuring.** A number obtained by counting has an infinite number of significant figures. There is no uncertainty when ten apples are in a bag whether it is 10.1 apples or 9.9 apples. Although we don't bother to write it, the correct number is 10.000000000000000000000.

Example: An apple weighs 50.12 g. The weight of 9 apples expressed in correct number of significant figures should be

9 X 50.12 g = 45.108 g = 45.11 g

- **Errors**. Find the absolute and percent relative uncertainty and express each answer with the correct significant figures.

For addition and subtraction, the uncertainty is obtained from the absolute uncertainties of individual terms.

e.g.1) $7.5(\pm 0.2) - 4.2(\pm 0.1) = ?$

answer = 3.3 ± error

$e = \sqrt{(0.2)^2 + (0.1)^2} = 0.2$

answer = 3.3 ± 0.2

e.g.2) $9.55\ (\pm 0.05) \times 0.016 (\pm 0.001) = ?$

0.1528 ± error

Convert absolute errors to % relative errors to estimate the number of significant figures.

9.55 (± 0.05 x 100/9.55) x 0.016 (± 0.001 x 100/0.016)

9.55 ((± 0.523%) x 0.016 (± 6.~~25~~%)

%error = $\sqrt{(0.523)^2 + (6.25)^2} = 6.30\ \%$

error = 6.30/100 x 0.1528 = 0.0096 = 0.01

answer = 0.15 ± 0.01

e.g.3)

MICROSOFT EXCEL

We will use Excel spreadsheet program to record, calculate, and graph numerical data. **Each final report should accompany a copy of the spreadsheet that was used**.

Example:

Given below is a spreadsheet from an experiment in which the weight was recorded each time after an addition of 1 milliliter of water from a burette to a beaker. The weight of the beaker was tared to zero at the beginning. The experiment was conducted in triplicate. Average weights and standard deviations were calculated using the functions available in Excel. In this example, slope gives the density of water. Always provide units when you report the slope and intercept. Together with the final lab report write up, a copy of the spreadsheet should be submitted as an appendix. Make sure the Row and Column numbers are included in your print out.

Mastering Excel takes time. Several useful tips are given below.

Excel Tip: How to Display multiple lines of text within a cell

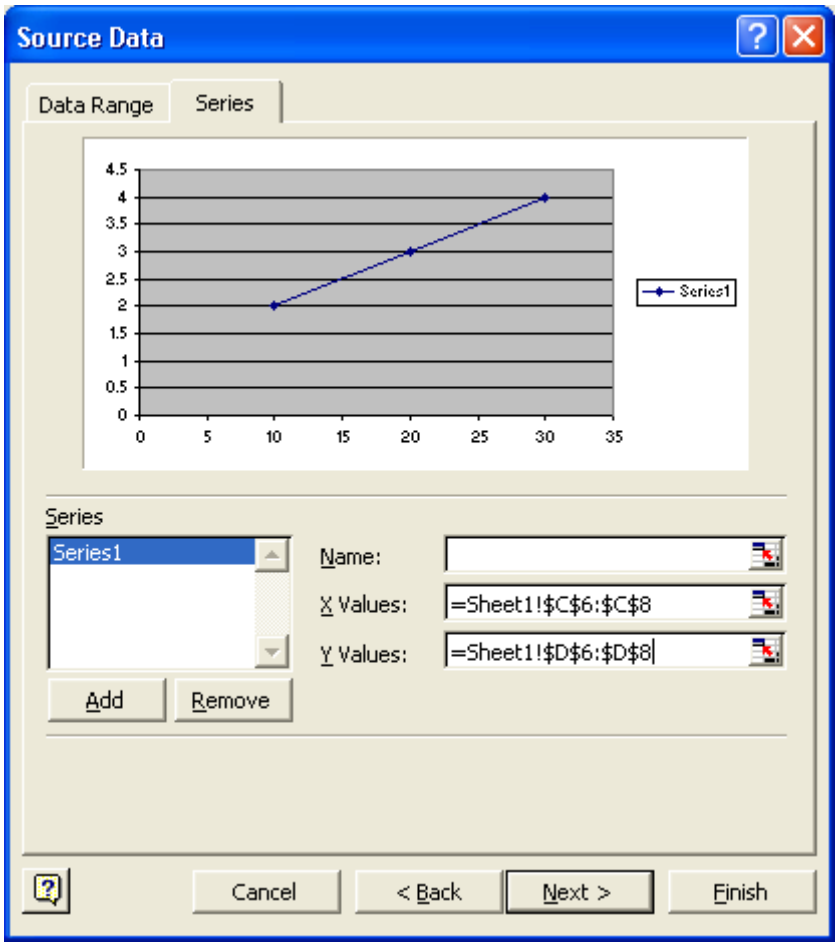
1. Select the cells you want to format.
2. On the **Format** menu, click **Cells**, and then click the **Alignment** tab.
3. Under **Text control**, select the **Wrap text** check box

Excel Tip: How to Display Greek symbols in axis labels.

1. First write Roman letters.
2. Click of the axis label. If it is the Y axis, click again. Select the letter to be changed. Click Format. Click "Selected Axis Title (or CTRL +1). Select SYMBOL as Font.

Excel Tip: How to Graph an X and Y Plot.

Data to be plotted must in two (or more) columns in the Spreadsheet. Click on an empty cell and go to the INSERT menu. Select CHART. For most of our purposes, the XY (scatter) is the most suitable selection. Many options will open up. Select the one in which data points are connected by a smooth curve. Click NEXT. Click on SERIES. Click on ADD. Place the cursor in the "X values:" box. Goto the spreadsheet and select the cells that contain data for X axis. Similarly, place the cursor in the "X values:" box, and delete what is displayed in the box. Place the cursor in the "Y values:" box and Goto the spreadsheet and select the cells that contain data for Y axis.



The above Figure shows that the X values for the plot are in cells C6:C8, and X values for the plot are in cells D6:D8.

The Data Range should now have "Columns" Radio Button selected. Click NEXT and follow the steps.

	A	B	C	D	E	F	G	H
1	**Weight of water per milliliter**							
2								
3		Weight of water recorded						
4		determination I	determination II	determination III	Average weight (g)	Average weight (g)	± SD	± SD
5	Volume (mL)							
6	1.00	1.0032	1.0046	1.2114	=AVERAGE(B6:D6)	1.0731	=STDEV(B6:D6)	0.1198
7	2.00	2.0150	2.2364	2.1009	=AVERAGE(B7:D7)	2.1174	=STDEV(B7:D7)	0.1116
8	3.00	3.0754	3.1307	2.9980	=AVERAGE(B8:D8)	3.0680	=STDEV(B8:D8)	0.0667
9	4.00	4.0065	4.1807	4.0097	=AVERAGE(B9:D9)	4.0656	=STDEV(B9:D9)	0.0997
10	5.00	5.0426	5.2438	5.1009	=AVERAGE(B10:D10)	5.1291	=STDEV(B10:D10)	0.1035
11	6.00	6.0496	6.2860	5.9905	=AVERAGE(B11:D11)	6.1087	=STDEV(B11:D11)	0.1564
12	7.00	7.0567	7.3283	6.9593	=AVERAGE(B12:D12)	7.1147	=STDEV(B12:D12)	0.1912
13	8.00	8.0637	8.3705	7.9281	=AVERAGE(B13:D13)	8.1208	=STDEV(B13:D13)	0.2267
14	9.00	9.0707	9.4128	8.8969	=AVERAGE(B14:D14)	9.1268	=STDEV(B14:D14)	0.2625
15								
16								
17								
18								
19								
20								
21								
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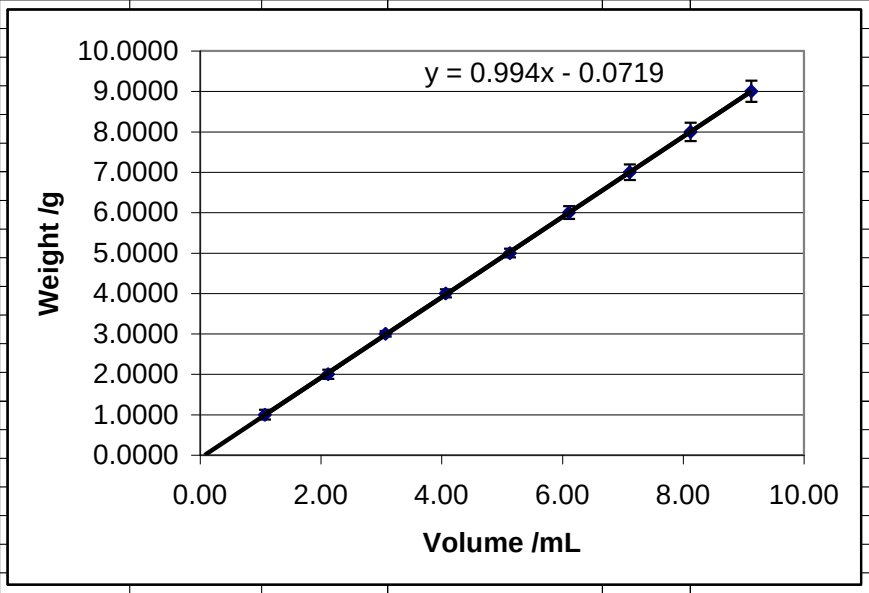


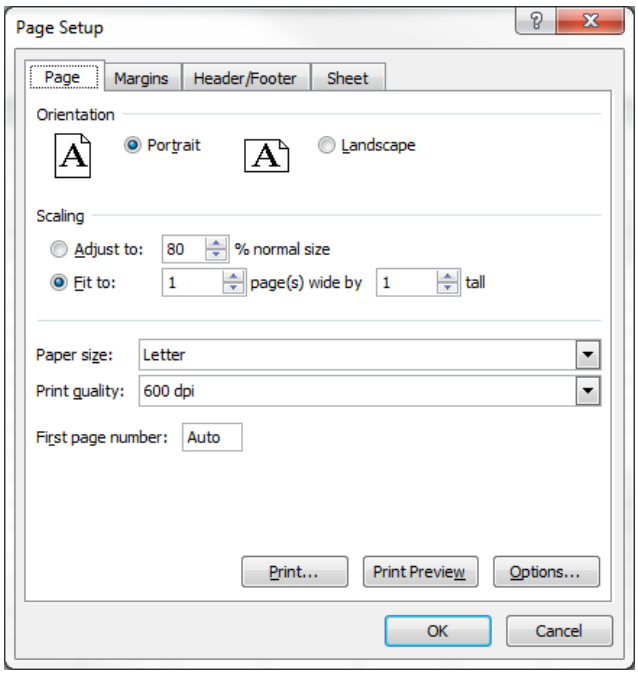
Fig. 1. An Excel Spreadsheet from the "weight of water per milliliter experiment."

The spreadsheet given above was pasted as "Paste Special…." "Picture (Enhanced Metafile)."

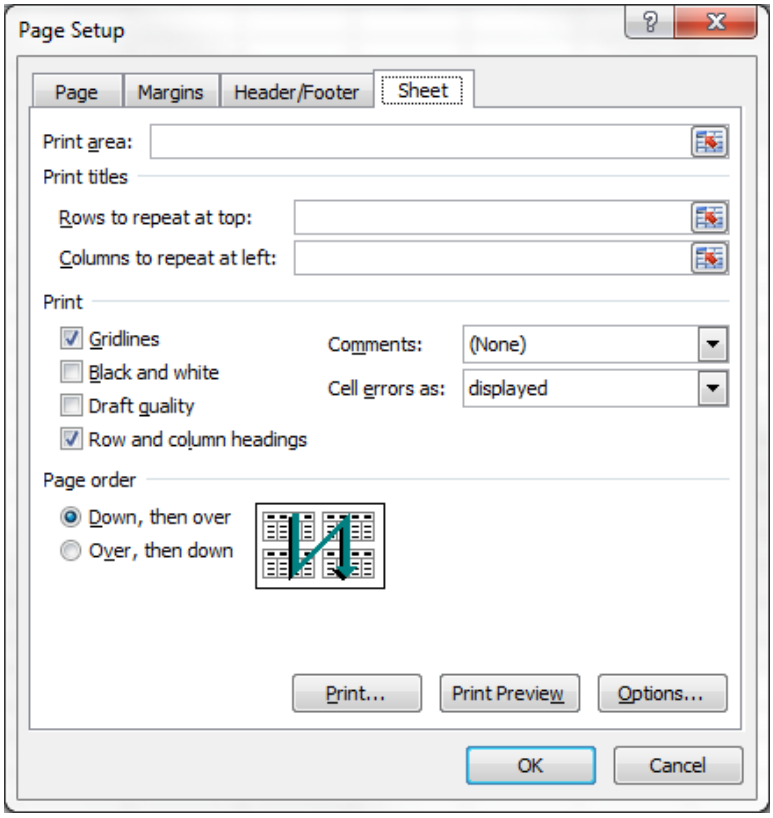
Excel Tip: How to Print an Excel Spread sheet with Column and Row labels.

1. From the *Page Layout* menu, click ***Page Setup... arrow***

The *Page Setup* dialog box appears.

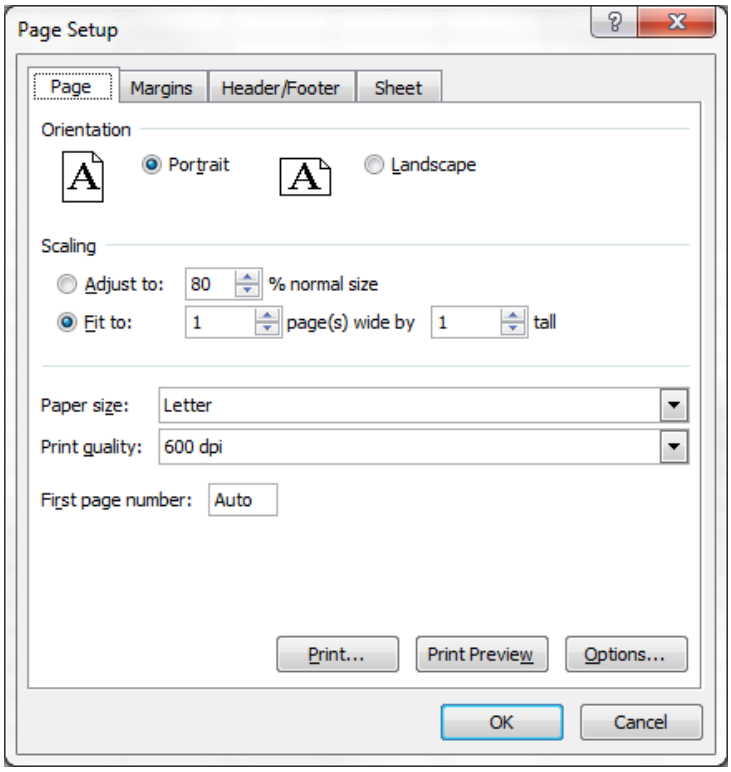


3. Select "Sheet."



3. Tick Mark “Gridlines” and “Row and column headings.”

4. You may also try the Fit to 1 Page Command in Page Setup.

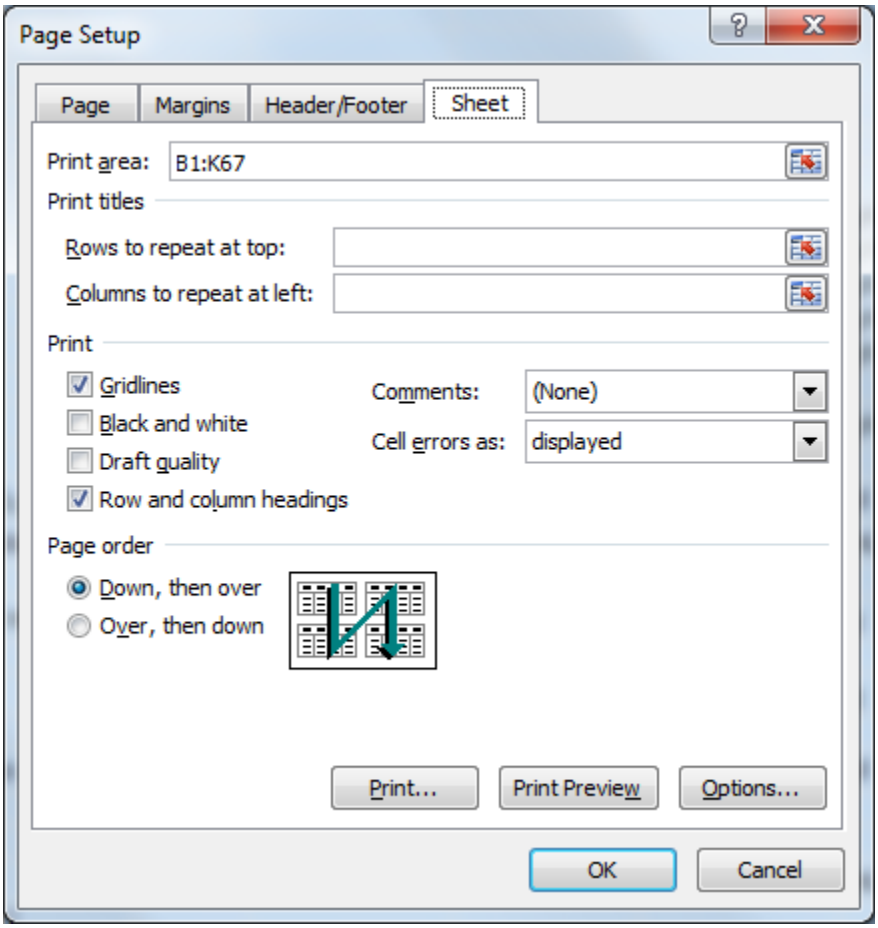


==========

You must learn how to add errors bars, insert Greek letters, superscripts and subscripts (Select the letters; Format; Cells, Subscript) in Excel charts.

Excel Tip: How to Print an Excel Spread sheet with Column and Row labels (Excel 2010)

1. Make sure a cell is selected (not a chart) on the spreadsheet.
2. Goto ***Pagelayout*** then to ***Page setup*** (click the slanted down arrow bottom in the bottom right-hand corner a new window should open).



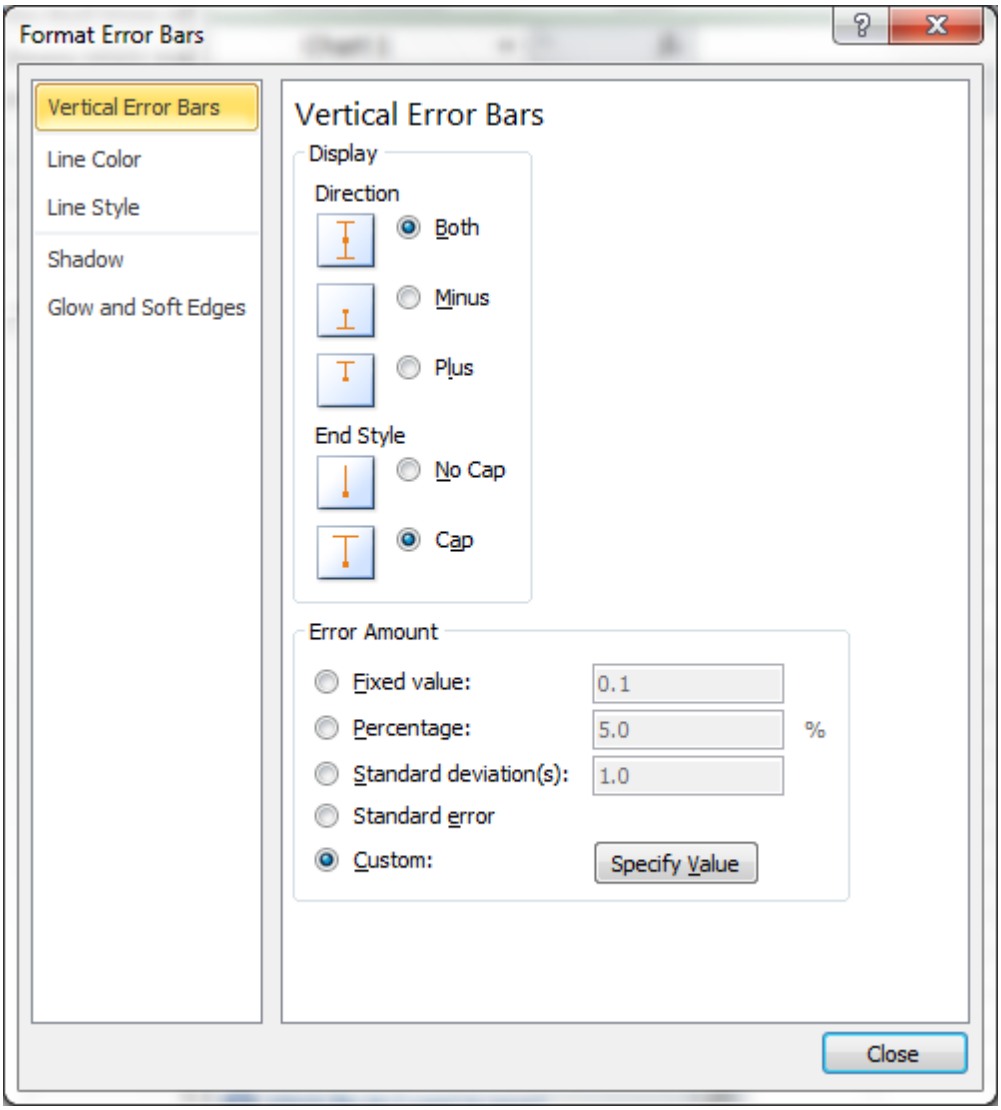
4. Select the ***Sheet*** tab
5. Check "Gridlines" and "Rows and volumn headings" boxes.
6. Do a "Print Preview" to see what you will get.

Excel Tip: How to Add Error Bars (Excel 2010).

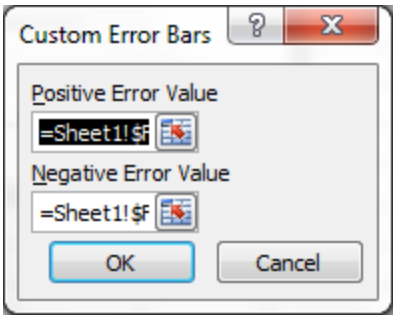
1. Plot the graph. Make sure that a ***Trendline*** has not been added to the chart.

2. Calculate the Standard Deviations of you dataset and add them to a new column (see Figure 1. Standard Deviations are in Column H).

3. **Click anywhere in the chart**. A new view will open for "Chart Tools." Chart Tools will be heighted and displayed (see top of the screen). Select the "Layout" tab. In the "Analysis" section, click the arrow pointing down. Click "more error bars Options…" A new window should open.



4. In "Display" box, check "Both."
5. In "Error Amount" box, check "Custom," and click "Specify value."



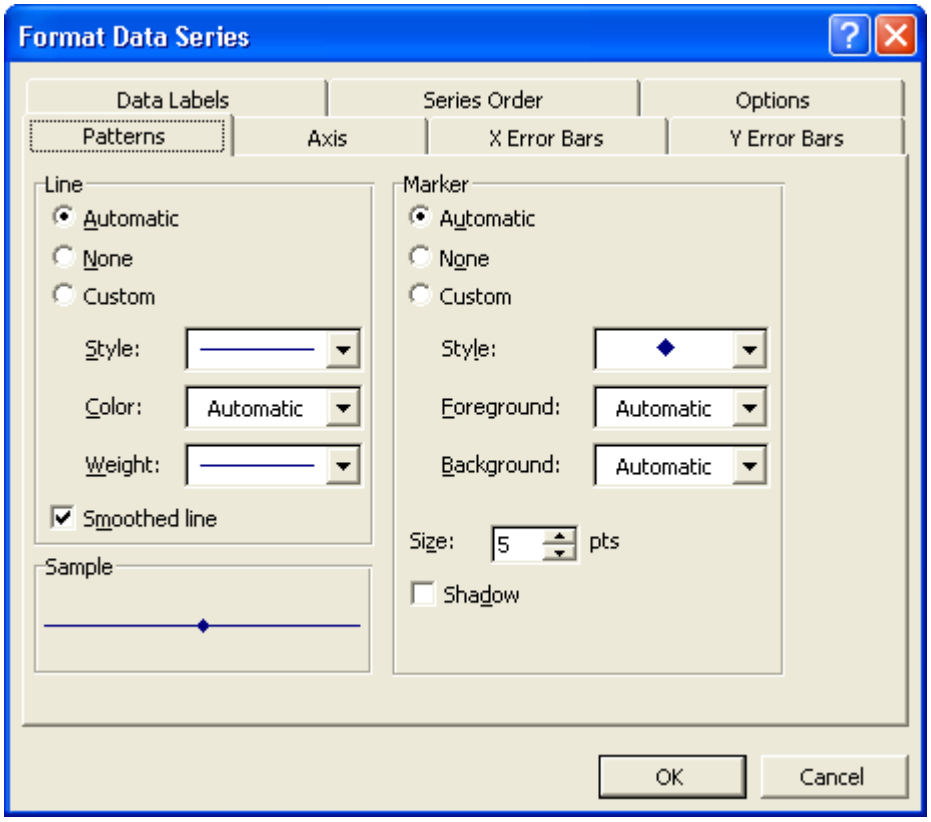
6. Click "Positive Error Value" box, and then select the column with SD values in your spreadsheet.
7. Click "Negative Error Value" box, and then select the column with SD values in your spreadsheet.

Excel Tip: How to Add Error Bars (Old versions of Excel).

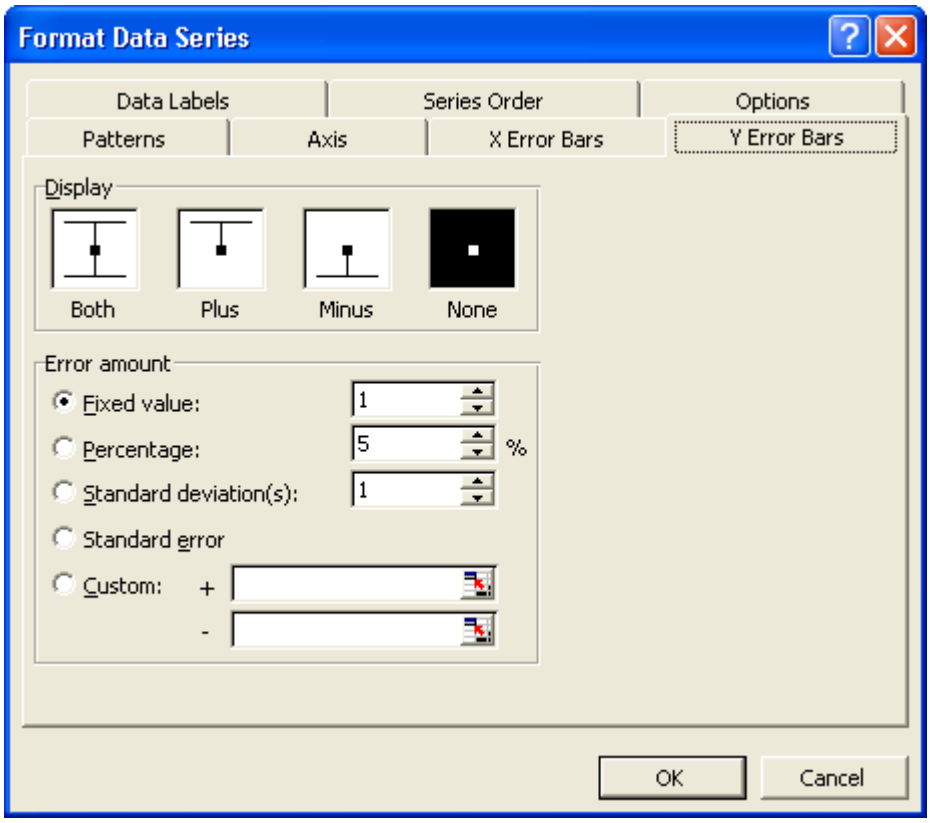
1. Plot the graph. Make sure that a ***Trendline*** has not been added to the chart.

2. Calculate the Standard Deviations and add them to a column (see Figure 1. Standard Deviations are in Column H).

3. Double click of a data point in the graph. The following window should open up.



4. Click on "Y Error Bars." The following window should open up.



7. Click on the Custom Radio button. Place the cursor in the "+ box," and select the column that contains the SD data. Then Place the cursor in the " - box," and select the column that contains the SD data (in this case the same column).

Solving polynomial equations

Many analytical problems involve solving quadratic (or polynomial equations of higher order such as cubic or quartic equations).

A quadratic equation is an equation than can be written in the following form:

$$ax^2 + bx + c = 0$$

where a is the coefficient of X^2, b is the coefficient of x, and c is a constant. This equation is called quadratic because the highest power of x is 2. The following formula, called the quadratic formula, allows calculation of values of x that satisfy the equation.

$$x = \frac{-b \pm \sqrt{b^2 - 4ac}}{2a}$$

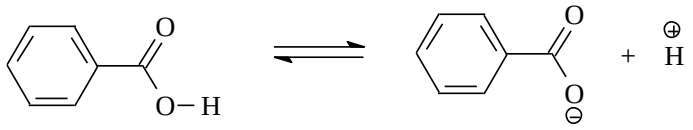
The ± sign indicates that there two solutions for this equation. However, for chemical calculations, a negative value is not meaningful. To obtain the positive root for x, using Excel, enter coefficients *a*, *b*, and *c* into cells, D11, D12, and D13, and correct formula into cells F16, F18, and F19.

	B	C	D	E	F
1					
2					
3	Solving quadratic equations				
4					
5	$ax^2 + bx + c = 0$				
6					
7					
8	$x = \frac{-b \pm \sqrt{b^2 - 4ac}}{2a}$				
9					
10					
11	a	=	3.000000		
12	b	=	6.000000		
13	c	=	-2.000000		
14					
15					
16	$\sqrt{b^2 - 4ac}$	=SQRT(D12^2-4*D11*D13)=			7.745967
17					
18	Positive root		x = (-D12 +F16)/2*D11=		0.290994
19	Negative root		x = (-D12 -F16)/2*D11=		-2.29099
20					
21					

Using SOLVER

Another excellent tool provided by Excel for solving polynomial equations is called **SOLVER**. [Look under **Tools** if you can see a button for **SOLVER.** If a button is not seen, goto **Tools**, and then **Add-Ins….**, and tick mark the **Solver Add-In** box (the computer might ask for the Microsoft Office installation CD).

Let's calculate the hydrogen ion concentration of a 0.0200 M benzoic acid solution. K_a of benzoic acid at 25 ^{0}C is 6.28 x 10^{-5} mol L^{-1}.



$$K_a = \frac{[H^+][A^-]}{[HA]} = 6.28 \times 10^{-5} \text{ mol L}^{-1}$$

	HA	⇌	H^+	+	A^-
Initial concentration (M)	0.0200		0		0
Change/mol	$-\alpha$		$+\alpha$		$+\alpha$
Total change (M)	-0.0200α		$+0.0200\alpha$		$+(0.0200)\alpha$
Equilibrium concentration (M)	$(0.0200) -(0.0200)\ \alpha =$ $0.0200(1-\alpha)$		0.0200α		0.0200α

$$K_a = \frac{[0.0200\,\alpha][0.0200\,\alpha]}{0.0200\,(1-\alpha)}\ \text{mol L}^{-1} = 6.28 \times 10^{-5}\ \text{mol L}^{-1}$$

$$\frac{0.0200\,\alpha^2}{(1-\alpha)}\ \text{mol L}^{-1} = 6.28 \times 10^{-5}\ \text{mol L}^{-1}$$

IF you multiply both sides of the equation by (1-α):

$$\frac{(1-\alpha)0.0200\,\alpha^2}{(1-\alpha)} = 6.28 \times 10^{-5}\ (1-\alpha)$$

$$0.0200\,\alpha^2 = 6.28 \times 10^{-5}\ (1-\alpha)$$

$$0.0200\,\alpha^2 = 6.28 \times 10^{-5} - 6.28 \times 10^{-5}\,\alpha$$

$$0.0200\,\alpha^2 + 6.28 \times 10^{-5}\,\alpha - 6.28 \times 10^{-5} = 0$$

$$ax^2 + bx + c = 0$$

Enter values for coefficients *a* and *b*, and constant *c* in three cells in an Excel spread sheet.

	A	B	C	D	E	F
1						
2		Solving quadratic equations suing SOLVER				
3						
4		$0.0200\ \alpha^2 + 6.28 \times 10^{-5}\ \alpha - 6.28 \times 10^{-5} = 0$				
5						
6		$ax^2 + bx + c = 0$				
7						
8		$x = \frac{-b \pm \sqrt{b^2 - 4ac}}{2a}$				
9						
10						
11						
12		a	=	2.0000E-02		
13		b	=	6.2800E-05		
14		c	=	-6.2800E-05		

Identify a cell as the “target cell,” in which the formula for x, which we want to set to zero is entered.

Identify another cell as the “answer cell” for x (SOLVER will enter the final answer to this cell)

D16 fx =D12*D17^2+D13*D17+D14

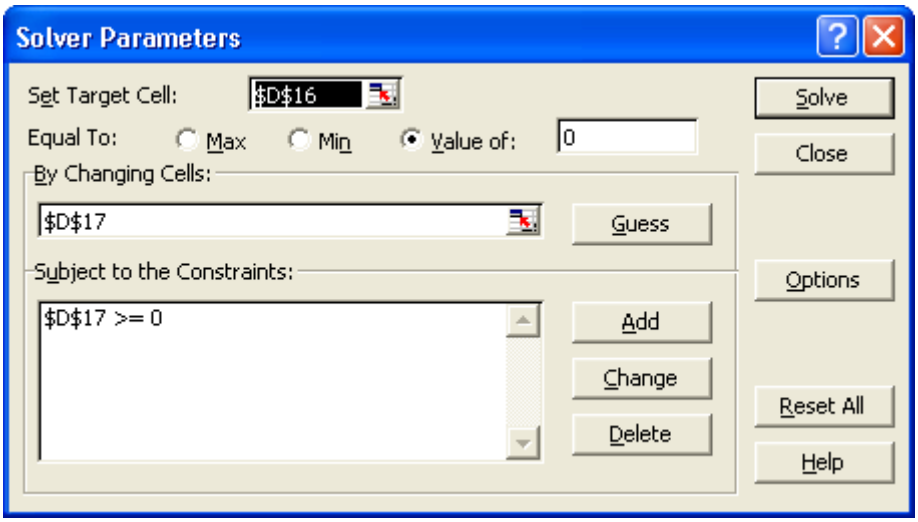
	A	B	C	D	E	F
1						
2		Solving quadratic equations suing SOLVER				
3						
4		$0.0200\ \alpha^2 + 6.28 \times 10^{-5}\ \alpha - 6.28 \times 10^{-5} = 0$				
5						
6		$ax^2 + bx + c = 0$				
7						
8		$x = \frac{-b \pm \sqrt{b^2 - 4ac}}{2a}$				
9						
10						
11						
12		a	=	2.0000E-02		
13		b	=	6.2800E-05		
14		c	=	-6.2800E-05		
15						
16		Equation		-6.00E-07		
17		α=		5.42E-02		
18						
19		H^+ conc		1.0844E-03		
20						
21						
22		pH	=	2.96		

From the above spreadsheet, you can see that we have selected D16 as the “target cell,” and D17 as the “answer cell.”

In D16, we have entered the function "=D12*D17^2+D13*D17+D14".

Goto **Solve**r from **Tools** menu.

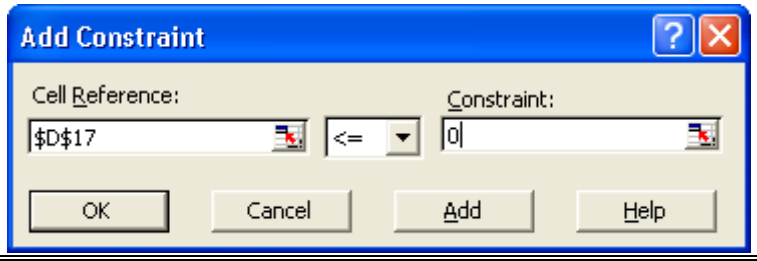
The **Solver Parameters** dialog box should open.



Place the cursor in the **Set Taget Cell** box, and click in the spreadsheet "**target cell**" that contains the calculation formula. For this example D16. Next, in the Equal to: line click the radio button **Value of:**, and enter 0 in the box.

Place the cursor in the **By Changing Cells:** box, and click in the spreadsheet "**answer cell.**" For this example D17.

Since the negative root is meaningless, we can instruct Excel to ignore it.



Matrix determinant and Cramer's Rule

We can use **Matrix determinant and Cramer's Rule** to solve multiple equations. For example, we wish to find the values of x and y of the following system (equations **1** and **2**).

$$x + 8y = 4 \quad \text{.....(1)}$$

$3x - y = -13$ .....(2).

Classically, we would solve equation **1** for x and apply that to equation **2** and find the answer.

$a_1x + b_1y = c_1$ .....(1)

$a_2x + b_2y = c_2$ .....(2)

To get rid of **y**, we multiply both sides of *equation 1* by b_2.

$$b_2[a_1x + b_1y = c_1]$$

Similarly, we multiply both sides of equation (2) by $-b_1$

$$-b_1[a_2x + b_2y = c_2]$$

Simplification gives us

$a_1b_2x + b_1b_2y = c_1b_2$ .....(3)

$-a_2b_1x - b_1b_2y = -c_2b_1$ .....(4)

Add equations (3) and (4) to get rid of *y*.

$(a_1b_2 - a_2b_1)x = c_1b_2 - c_2b_1$

$$x = \frac{c_1b_2 - c_2b_1}{a_1b_2 - a_2b_1} \quad \text{.....(5)}$$

Similarly, we can get rid of *x* and find a value for *y*.

$$y = \frac{a_1c_2 - a_2c_1}{a_1b_2 - a_2b_1} \quad \text{.....(6)}$$

However, this procedure becomes too difficult when there are many equations and many unknowns. Matrix algebra provides us an easier method.

How can we represent $a_1b_2 - a_2b_1$ by a matrix.

$$\begin{bmatrix} a_1 & b_1 \\ a_2 & b_2 \end{bmatrix} = a_1 b_2 - a_2 b_1$$

This is a 2 by 2 matrix.

==========

A rectangular array of numbers such as

$$A = \begin{bmatrix} a_{11} & a_{12} & a_{13} \\ a_{21} & a_{11} & a_{11} \\ a_{31} & a_{31} & a_{31} \end{bmatrix} = \begin{bmatrix} 2 & 1 & 3 \\ 3 & -1 & -2 \\ 2 & 3 & 1 \end{bmatrix}$$

is called a matrix. This is a 3 by 3 matrix because it has three rows and three columns (An *m* by *n* matrix has *m* rows and *n* columns). In fact, this is a **square matrix** since it has the same number of rows and columns. It is a **matrix of order 3** since it has three rows and three columns. Each square matrix is associated with a number called the **determinant**.

$$D = \det\ A = \det \begin{bmatrix} a_{11} & a_{12} & a_{13} \\ a_{21} & a_{22} & a_{23} \\ a_{31} & a_{32} & a_{33} \end{bmatrix} =$$

Systems of three equations and three unknowns, x , y, and z, can be dealt in the same way. If $D \neq 0$, the system has a unique solution, given by Cramer's rule:

$$x = \frac{1}{\mathrm{D}} \begin{bmatrix} b_1 & a_{12} & a_{13} \\ b_2 & a_{22} & a_{23} \\ b_3 & a_{32} & a_{33} \end{bmatrix}$$

$$y = \frac{1}{\mathrm{D}} \begin{bmatrix} a_{11} & b_1 & a_{13} \\ a_{21} & b_2 & a_{23} \\ a_{31} & b_3 & a_{33} \end{bmatrix}$$

$$z = \frac{1}{\mathrm{D}} \begin{bmatrix} a_{11} & a_{12} & b_1 \\ a_{21} & a_{22} & b_3 \\ a_{31} & a_{32} & b_3 \end{bmatrix}$$

Example:

Evaluate the following determinant.

$$\begin{bmatrix} 2 & 1 & 3 \\ 3 & -1 & -2 \\ 2 & 3 & 1 \end{bmatrix}$$

2	1	3	2	1	
3	-1	-2	3	-1	
2	3	1	2	3	
			-2	-4	27

			-6	-12	3
2	1	3	2	1	
3	-1	-2	3	-1	
2	3	1	2	3	
			-2	-4	27

Change these signs

Keep these signs

Add them up to find the determinant

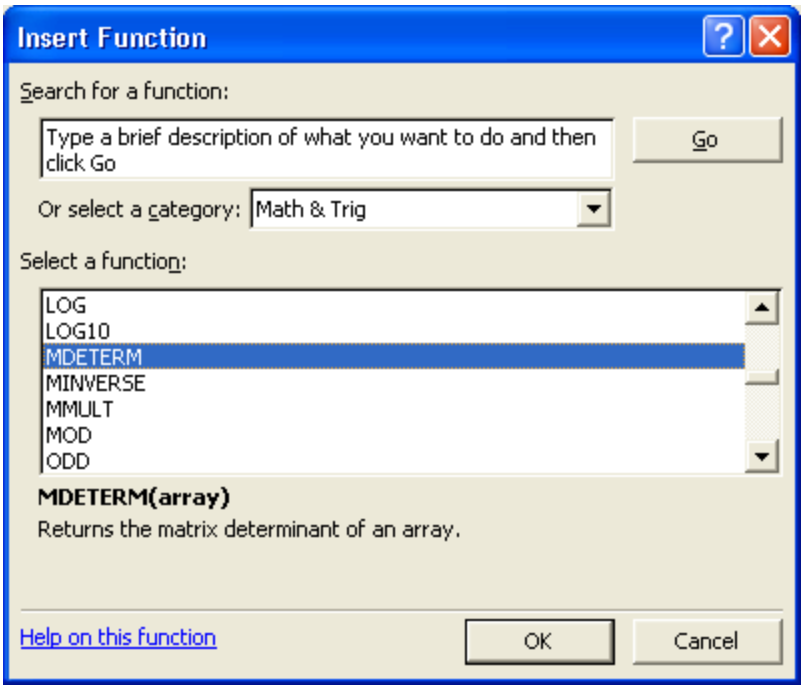
Det A = -(-6) - (-12) -3 + (-2) + (-4) + 27 = 36

Excel provides such matrix calculations at a click of a button.

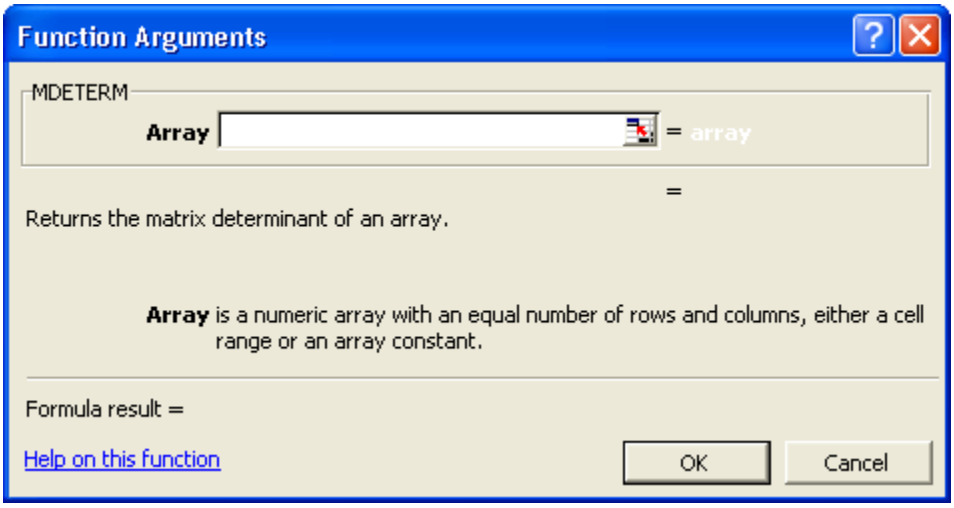
For example.

	A	B	C
1	2	1	3
2	3	-1	-2
3	2	3	1

Click on any empty cell, click on "f_x" in tool bar, and INSERT FUNCTION window will open.



Select the function MDETERM, and say OK.



Select the cells containing your matrix and say OK.

Alternatively, you may just type `=MDETERM(A1:C3) in any cell.` The matrix determinant is a number derived from the values in the array `A1:C3`.

For a three-row, three-column array, A1:C3, the determinant is defined as:

`MDETERM(A1:C3)` equals
`A1*(B2*C3-B3*C2) + A2*(B3*C1-B1*C3) + A3*(B1*C2-B2*C1)`

Cramer's Rule

If $D \neq 0$, the following system has a unique solution.

$$D = \det A = \det \begin{bmatrix} a_{11} & a_{12} \\ a_{21} & a_{22} \end{bmatrix} \neq 0$$

$a_{11}x + a_{12}y = b_1$ .....(1)

$a_{21}x + a_{22}y = b_2$ .....(2)

Equations **1** and **2** have two unknowns, x and y. Cramer's rule states that x and y may be found from the formulas

$$x = \frac{1}{\mathrm{D}} \begin{bmatrix} b_1 & a_{12} \\ b_2 & a_{22} \end{bmatrix}$$

$$y = \frac{1}{\mathrm{D}} \begin{bmatrix} a_{11} & b_1 \\ a_{21} & b_2 \end{bmatrix}$$

This is best understood by working an example.

$x + 8y = 4$ .....(3)

$3x - y = -13$ .....(4) Solve this system and find values of x and y.

Answer:

First find the determinant of the coefficient matrix

$$D = \begin{bmatrix} 1 & 8 \\ 3 & -1 \end{bmatrix} = -1 - (8 \times 3) = -25$$

$$x = \frac{1}{\mathrm{D}} \begin{bmatrix} 4 & 8 \\ -13 & -1 \end{bmatrix} = -4 - (-13 * 8) / -25 = -4$$

$$y = \frac{1}{\mathrm{D}} \begin{bmatrix} 1 & 4 \\ 3 & -13 \end{bmatrix} = 1$$

Pre Lab Exercises:

1) Find the following 3rd order determinant.

$$D = \begin{vmatrix} 4 & -6 & 0 \\ 0 & 4 & -1 \\ 1 & 2 & 1 \end{vmatrix}$$ Answer = 30

2) Find the following 4th order determinant (use Excel).

$$D = \begin{vmatrix} 4 & -6 & 0 & 1 \\ 0 & 4 & -1 & 2 \\ 1 & 2 & 2 & -2 \\ 2 & -2 & 1 & -1 \end{vmatrix}$$ Answer = -12

3) Solve the following systems of equations by Cramer's rule.
   a. 5x – 2y = 11 Answer : x = 5, y = 7
      2x + 3y = 31

4) Solve the following systems of equations by Cramer's rule.
   a. 2x + 1y -2z = 10 Answer: x = 3, y = 2, z = -1
      3x + 2y -z = 14
      x + 3y +z = 8

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Logarithms

It is important to know the following

$X = 10^z$ $\log_{10} x = z$

$100 = 10^2$ $\log_{10} 100 = 2$

Log (a * b) = log a + log b

Log (a /b) = log a - log b

Log (a /b) = - log (b/a)

Log 1 = 0

$$A = \log\left[\frac{I_0}{I}\right] = -\log\left[\frac{I}{I_0}\right] = -\log T = \log\frac{1}{T}$$

Logarithms

Natural logarithms (ln) are based on the number e which is equal to 2,718281….

ln 10 = 2.303

You must be able to convert between ln x and $\log_{10} x$

Logarithms and Antilogarithms

$$10^{-4} = \frac{1}{10^4} = \frac{1}{10000} = 0.0001$$

number	base 10 LOG	ln
1	0	0
10	1	2.302585
100	2	4.60517
339	2.530	5.826
1.00E-07	-7.00	-16.1181
1.50E-07	-6.82	-15.71263

antilog (-6.82) $= 10^{-6.82} = 10^{-6} \times 10^{0.18} = 0.15 \times 10^{-6} = 1.5 \times 10^{-7}$

Example 1: What is the pH of a 1.5 X 10^{-7} M H^+ ion solution?

pH = -log(H^+ ion concentration)

pH = -log(1.5 X 10^{-7})

In EXCEL you may write a cell function, =-LOG10(1.5E-7)

Which will return the answer 6.82.

Example 2: What is the H^+ ion concentration of a solution pH of a 6.82?

In EXCEL you may write a cell function, =10^-6.82

Which will return the answer 1.5E-07

Example 3:

Solve the following equation

$$0.227 = -\frac{0.059}{2}\log\frac{[x]^2[x]^2}{[0.5]}$$

$$0.227\left[\frac{2}{0.059}\right] = -\log\frac{[x]^2[x]^2}{[0.5]}$$

$$7.6949 = -\log\frac{[x]^2[x]^2}{[0.5]}$$

$$10^{-7.6949} = \frac{[x]^2[x]^2}{[0.5]}$$

$$2.0187\ \text{x}\,10^{-8} = \frac{[x]^2[x]^2}{[0.5]}$$

$$1.0094\ \text{x}\,10^{-8} = [x]^4$$

$$\sqrt[4]{1.0094\ \text{x}\,10^{-8}} = x$$

x = 0.01

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To add Mathematical Equations to Your Research Reports Use Microsoft Equation Editor

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You can insert a mathematical equation into Word, Excel, or PowerPoint, from **Insert** menu. However, you may have to install the program from you **Office** CD.

1. Insert your copy of **Office** CD in CD-ROM drive.
2. Open the **`Control Panel`**.
3. Double-click the **`Add/Remove Programs`** icon.
4. In the white box, locate the Microsoft Office entry that corresponds to what you have installed and click once to highlight it. For example, if you have Office XP, locate **`Microsoft Office XP Professional`** and select it.
5. Click the **`Change`** button. When the `Office installation` window opens, select **`Add or Remove Features`**, then click **`Next`**.
6. Next to **`Office Tools`**, click the **`+`** (plus sign) once. Click **`Equation Editor`**, and select **`Run from Computer`**.
7. Click **`Continue`**. When Equation Editor has finished installing, click **`OK`**.

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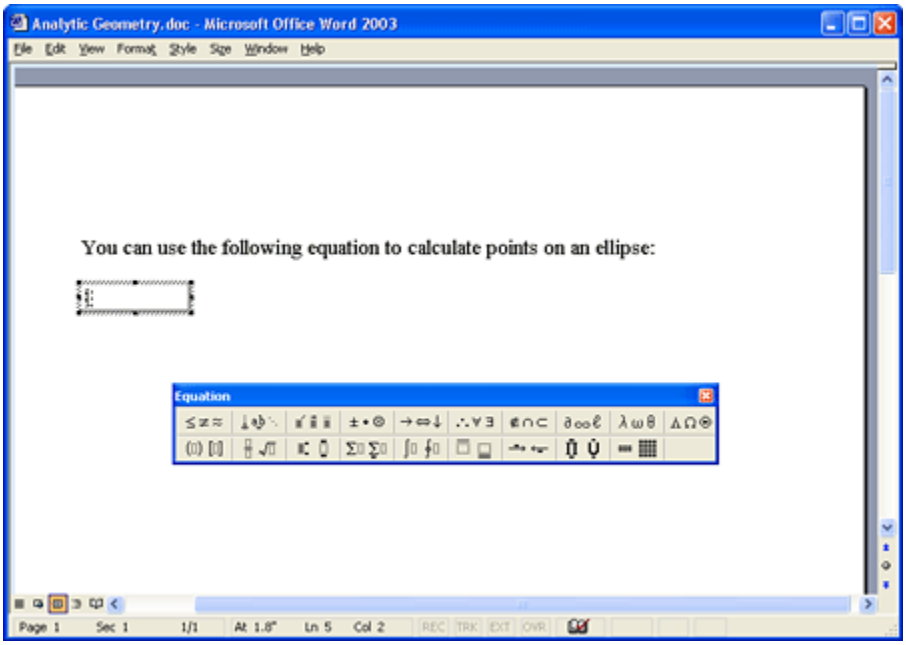
To enter a mathematical equation, do the following:

1. On the **Insert** menu, click **Object**.
2. In the working area into your document, and the **Equation** toolbar and menus will appear within the Office program's window.



The equation you insert is an embedded object created by the Equation Editor program.