Chemistry 2030 / 2130 Laboratory Manual
Summer 2017

Senior Laboratory Instructor
Richard Bartholomew, UA 3071
richard.bartholomew@uoit.ca

Table of Contents

Chemistry 2130 students will do the Excel Tutorial and experiments 1, 5, 7, 8

ACKNOWLEDGEMENTS

Over the years this manual has evolved (and improved!) thanks to the hard work and helpful suggestions of a number of people including technicians, teaching assistants and students. These include:

Michael Allison - the technician who started it all and corrected many mistakes.
Laura Benninger
Patrick Edge
Shannon Hill
Clayton Jakins - who made many helpful suggestions and is the technician who looks after it all now.
Shumail Kamal
Stephanie Mavilla (one of the "Tigers")
Lori Van Belle (another one of the "Tigers"!) - who tested many of the experiments and found the mistakes.
Ömer Yükseker

As all authors say, any mistakes that remain are entirely my own!

LABORATORY CALENDAR (SUMMER, 2017)

After completing the last experiment you must "check out". You must ensure all the equipment that is supposed to be in your locker is in the locker (no more, no less!) and that it is clean and in good repair.

Chemistry 2030

May 9	Laboratory Skills Tutorial / Excel Tutorial
May 16	1 - Statistical Analysis of Laboratory Technique
May 23	2 - Statistical Evaluation of Indicators
May 30	3 - The Importance of Sampling in Chemical Analysis
June 6	4 - The Determination of $[Ca^{2+}]$ by an EDTA Titration
June 13	5 - Iodometric Titration of Vitamin C
June 20	"Reading" Week - no experiments
June 27	6 - Mass Titration - Variation on an Old Theme
July 4	7 - Spectrophotometric Analysis of Glucose
July 11	8 - Determination of pK_a of an Acid - Base Indicator
July 18	9 - Gran Titration to Determine Concentration of Acetic Acid
July 25	All laboratory reports due

May 9, 16	Laboratory Skills Tutorial / Excel Tutorial
May 23, 30	1 - Statistical Analysis of Laboratory Technique
June 6, 13	5 - Iodometric Titration of Vitamin C
June 27, July 4	7 - Spectrophotometric Analysis of Glucose
July 11, 18	8 - Determination of the pK_a of an Acid - Base Indicator
July 18, 25	All laboratory reports due

PROPOSED WORK PLAN FOR CHEMISTRY 2030

Here is is a proposed plan for organizing your time in the laboratory. It is only a proposal; if you have one of your own, let us hear it! A big part of your success as a "bench" chemist will come from your ability to organize your time and work under pressure.

Experiment	Preparatory work
2 - Statistical Evaluation of Indicators	Dry ~0.6 g of $CaCO_3$ for experiment 4.
3 - The Importance of Sampling in Chemical Analysis	Prepare standard solution of $CaCl_2$ for experiment 4.
4 - The Determination of $[Ca^{2+}]$ by an EDTA Titration	Boil ~600 mL of deionized water and dry 1.0 g of KIO_3 for experiment 5.
5 - Iodometric Titration of Vitamin C	Dry potassium hydrogen phthalate for experiment 6.
	Ensure you have 9 clean 20 x 150 mm test tubes for experiment 7.
6 - Mass Titration - Variation on an Old Theme	Prepare glucose working standard for experiment 7.
8 - Determination of the pK_a of an Acid - Base Indicator	Boil 600 mL of deionized water and dry ~1.5 g of potassium hydrogen phthalate for experiment 9.

Special Notes for Chemistry 2030 Students

The Laboratory Skills Tutorial and Experiments 1 - 3 are intended for you to learn and practise the laboratory skills you will need to succeed in a "wet" analytical chemistry laboratory.

You should note that the marks for experiments 4 - 7 and 9 will be based largely on the accuracy and precision of your results.

Your marks will depend heavily on your results and this, in turn, depends on the condition of your glassware (particularly cleanliness). Consequently, you will each be assigned your own locker with your own glassware. You are solely responsible for maintaining the glassware in your locker and only you will have access to it. Be sure your locker is locked at the end of your laboratory period. Each locker is also assigned a set of pipettes and a burette for use during the semester. This glassware is labelled with the locker number and has a specific location (also labelled with the locker number) for storage. Be sure to return the glassware to the correct location so that you can easily find it. Check, periodically, that the label is still visible on the glassware. Do **NOT** use the glassware of another student.

For the last half of the course you may have to work on several experiments at the same time in order to complete all of the experiments in your allotted laboratory periods this semester. Such things as drying primary standards and solution preparation may have to be spread over several laboratory periods. It is expected that different students may be working on different experiments at different times. Students should do as much preparation as possible (e.g. calculations) before coming to the laboratory in order to use laboratory time efficiently. The normal schedule for laboratories will have 10 periods in a semester.

Lockers for Chemistry 2130

Chemistry 2130 students will have to share their lockers with at least one other student (and perhaps more than one). It is the student's responsibility to ensure that the glassware and equipment in the locker is clean and undamaged at the end of the laboratory period. At the end of the period, either the teaching assistant or the technician will check the locker to ensure everything is in order. Your notebook will not be signed until the locker, equipment and workspace are in a satisfactory condition.

LEARNING OBJECTIVES FOR CHEM 2030U / 2130U LABORATORIES

At the end of the laboratory portion of CHEM 2030U / 2130U students should accomplish the following:

"Theoretical"

1. Students should know how to determine if "real" (i.e. statistically significant) differences exist between measured quantities or between measured quantities and standard values.

2. Students should be able to perform all the calculations associated with direct titrations and back titrations.

3. Students should be able to perform all calculations associated with solution preparation. For example, they should be able to determine the required mass of solute for a given concentration and volume (including mass of salt required to achieve a prescribed concentration of one of the ions), concentration from mass of solute and volume of solution. They should be able to account for dilution.

4. Students should be able to do all calculations associated with buffer preparation.

"Practical"

1. Students should be capable of performing basic laboratory calculations, creating proper graphs, and doing regression analyses using Excel.

2. Students should know how to properly prepare and use transfer and Mohr pipettes and know the differences between the two.

3. Students should know how to properly prepare a standard solution and the importance of good mixing.

4. Students should know proper weighing techniques (particularly "weight-by-difference"). They should know the differences between the open-pan balance and the analytical balance and when to use each.

5. Students should know how to prepare a burette and how to titrate properly.

6. Students should be able choose equipment appropriately for efficient conduct of experiments. For example, they should know when a graduated cylinder can be used to transfer 10 mL of liquid and when a pipette is necessary to transfer 10.00 mL of liquid.

"Safety"

1. Students should know how to properly dress for the laboratory: safety glasses (or goggles), lab coat, proper shoes, etc.

2. Students should know the basics of handling hazardous waste: e.g., where chemicals should be disposed, the need for segregation of waste, proper filling of waste vessels.

3. Students should know the basic principles of handling strong acids and bases.

4. Students should recognize that glassware and its improper use may pose one of the greatest hazards in a chemistry laboratory. Students should be "mindful" when handling and cleaning

glassware and be always aware of the potential dangers (e.g., no shaking of glassware to remove solutions, no attempted drying / draining of glassware when glassware need not be dry).

LABORATORY ATTENDANCE AND REPORT SUBMISSION POLICIES

Attendance at laboratory periods and submission of laboratory reports are compulsory. Failure to attend a laboratory period or to submit the report may result in a grade of zero. A student who fails to submit more than **TWO (2)** laboratory reports will not receive credit for the laboratory portion of the course. This may result in failure of the course. If you miss a laboratory period for any reason, please read the "Missed Experiments" section of the laboratory manual and follow the instructions there.

Students must arrive on time at the start of the laboratory period to receive instructions about the safe conduct of the experiments. Students who arrive more than 10 minutes late will not be allowed access to the laboratory. Students arriving less than 10 minutes late to a laboratory period will be admitted at the discretion of the teaching assistant. If you are forbidden entry to a laboratory period because of lateness, please read the "Missed Experiments" section of the laboratory manual and follow the instructions there.

Details about laboratory reports (format, deadlines, etc.) are given in the "Laboratory Reports" section of the this manual.

When you submit your laboratory report be sure to have the teaching assistant (or whomever receives your report) sign your "Laboratory Report Receipt Sheet"; it is your proof that you submitted your report and if the report is lost, you can use the receipt sheet to demonstrate that you submitted the report. In circumstances in which the report is lost and the Laboratory Report Receipt Sheet has been signed, the final laboratory grade will be calculated based on the marks for the remaining laboratory reports. If the Laboratory Report Receipt Sheet has not been signed and the report is lost, a mark of zero will be recorded. It is *your* responsibility to have the receipt sheet

signed.

Your marked laboratory report will normally be returned to you in the laboratory period following the submission of the report. If your report is not returned to you at this time, confirm with your teaching assistant that (s)he has it. This is particularly important if you submitted your report to someone other than your teaching assistant. If you suspect your report is missing, you must alert the senior laboratory instructor (richard.bartholomew@uoit.ca) no more than **three (3)** working days after the laboratory period in which the report should have been returned to you. Failure to inform the senior laboratory instructor of the missing report by this deadline may result in a grade of zero for the laboratory report.

If you have suffered *exceptional* extenuating circumstances (e.g., grave personal misfortune, acute mental health problems) which may prevent you from submitting a laboratory report on time, you must contact the senior laboratory instructor no more than **five (5)** working days after the due date for the laboratory report in order to be considered for a special accommodation. Documentation may be required. All applications for such accommodations **MUST** be made by **e-mail** to the senior laboratory instructor: richard.bartholomew@uoit.ca. No accommodations will be granted for late applications.

SAFETY RULES

Safety in the laboratory is of the utmost importance to your instructors and it must be to you, as well. Everybody's safety depends on each student adhering to the safety rules and procedures outlined in this manual. For first-year students, a YouTube video is available for you to watch: Laboratory Safety Video (which may be a useful reminder to upper year students).

The following rules must be obeyed and will be rigorously enforced.

1. **ALL** students must wear eye protection. For people with eyeglasses it must be worn over regular glasses. This rule will be vigorously enforced.

2. Contact lenses should not be worn in the chemistry laboratory.

3. All students must wear a lab coat. A 100% cotton lab coat is best. Lab coats protect skin and clothing from chemicals. The front of the lab coat should be buttoned while working in the laboratory. Cotton generally provides better protection from fire than synthetic fibres.

4. Open shoes or sandals are forbidden. They expose your feet to spilled chemicals. Footwear must completely enclose the foot from ankle to toe.

5. Loose or bulky clothing presents a hazard and should not be worn in the laboratory.

6. Clothing that exposes large areas of the body (e.g. shorts, tank tops) must not be worn in the laboratory.

7. Long hair must be tied back. The simple rule: if it can be tied back, it must be tied back!

8. Aisles must be kept clear of boots, coats, knapsacks, etc.

9. Students must know the locations of:

   a) fire extinguishers
   b) eye wash stations
   c) emergency showers
   d) emergency exits
   e) fire blanket
   f) fire alarms

10. The eyewash stations, shower, fire blanket and fire extinguishers must not be obstructed.

11. Smoking, eating and drinking are all strictly forbidden in the laboratory. Neither food nor drink may be brought into the laboratory. This includes water bottles or canteens. These must be left outside the laboratory.

12. No laboratory may be started without an instructor present. Unauthorized experiments are strictly forbidden. Experiments must not be left unattended.

13. Never put broken glass in the regular garbage. To do so presents risks to cleaning staff. Broken glass should be swept up with a dust pan and brush and disposed in the receptacle for broken glass. <u>ONLY</u> broken glass should be placed in this container.

14. When diluting acid be sure to add the acid to the water. Add the acid slowly and with plenty of stirring. Diluting acids generates huge amounts of heat (it is a very exothermic process). Keeping the water in excess allows this heat to be more effectively dissipated. If the heat is not adequately dissipated, the rapid heating may cause the solution to be ejected from the

beaker, injuring the experimenter. If the acid is added to the water, the ejected solution will be comparatively dilute and therefore less dangerous.

15. Organic solvents and inflammable or pyrophoric (igniting spontaneously when exposed to air) reagents require special handling. They should be used only in a fume hood.

16. **NEVER, EVER** pipette by mouth. Safety bulbs are provided for pipetting. When pipetting by mouth, it is very easy to lift the tip of the pipette above the level of the liquid being pipetted As a result, solution will enter the mouth.

17. Never point the mouth of a test tube at yourself or others. This is especially true if the test tube is being heated.

18. **NEVER** taste chemicals or solutions. Should you get any chemicals in your mouth, do not swallow. Rinse your mouth thoroughly and then consult an instructor.

19. If you must smell a chemical, waft the vapours toward your face with your hand. Do not place the container directly under your nose - you may be overcome by the gas.

NOTE: Students who arrive for laboratories inappropriately addressed may be forbidden from performing the experiment. Students who violate the safety rules may be dismissed from the laboratory period. In such cases the student will be given a grade of zero for the laboratory report.

SAFETY PROCEDURES

Fire

Fire is one of the most serious problems that may be faced in the chemistry laboratory. For no other safety issue is the adage "an ounce of prevention is worth a pound of cure" more applicable.

For all fires, if time permits, turn off all services (burners, hot plates, water, etc.).

In the event of a small fire, use the fire extinguisher. Remove the pin by twisting and pulling it out. Direct the nozzle of the extinguisher at the base of the fire and squeeze the trigger. Ensure that at all times you are between the fire and your escape route. Small fires can rapidly and easily become large fires. If the fire cannot be safely extinguished in 30 s, leave the room.

For large fires leave the room and pull the fire alarm. If in doubt, leave the room and pull the fire alarm. The more time people have to escape the more likely they will be successful. The last person out of the room must close the door – this is the single most effective way to slow the spread of fire. Leave the building by the nearest exit and move well away from the building. The teaching assistant should call emergency services and give all necessary details to the fire crews.

Clothing on Fire

DO NOT RUN! Wrap the victim in the fire blanket or use a lab coat to smother the flames. "Drop and Roll" is effective. The emergency shower can be used. Have someone call the emergency number and get medical assistance.

Fire Alarm / Building Evacuation

UOIT uses a "two stage" fire alarm system. The first stage is a warning that there may be an emergency in the building. In the first stage the fire alarm rings at a rate of 30 rings per minute. At the first stage of the alarm, stop all your experiments when it is safe and convenient to do so and prepare to leave the building. Await further instructions. If the fire alarm progresses to the second stage (louder, more frequent alarm), immediately stop all experiments. Turn off all services (gas, electricity, water, etc.). Before leaving the laboratory make sure the hallway is free of smoke and fire. If it is, leave the laboratory and exit the building by the quickest route (see below). Close the door to the laboratory. Once outside, move away from the building and assemble at the western end of the first floor atrium of UB. If UB is closed, meet in the car park immediately to the east of UA. Your TA will do a headcount to ensure everyone is out. Do **NOT** wander away without telling your TA! Under no circumstances should you return to the building until told by the fire department that it is safe.

University authorities may order the evacuation of university buildings for reasons other than for fire. Under such circumstances you should follow the instructions from security without delay.

Evacuation Routes from Laboratories

Room	Primary Escape	Secondary Escape
Instrument Room	Turn right, descend staircase at southeast corner of building. Exit building by southeast entrance. Assemble in Founders 1 car park.	Turn left, descend staircase at the northeast corner of the building. Exit by the north doors. Assemble in UB atrium.

Room	Primary Escape	Secondary Escape
UA 3420	Turn left, descend staircase at northwest corner of atrium. Exit building by north doors.	Turn right, then left along southern corridor. Descend staircase at southeast corner of building. Exit building by doors at southeast corner.
UA 3480	Turn left, descend staircase at northwest corner of atrium. Exit building by north doors.	Go straight along southern corridor. Descend staircase at southeast corner of building. Exit building by doors at southeast corner.
UA 3520	Turn right, descend staircase at southeast corner of building. Exit building by southeast entrance. Assemble in Founders 1 car park.	Turn left, then right along western corridor. Descend staircase at northwest corner of atrium. Exit building by north doors. Assemble in UB atrium.
UA 3680	Turn right, descend staircase at the northeast corner of the building. Exit by the north doors. Assemble in UB atrium.	Turn left, descend staircase at southeast corner of building. Exit building by southeast entrance. Assemble in Founders 1 car park.

Medical Situations

UOIT maintains a service called the Campus Emergency Response Team (CERT). CERT is staffed by volunteers who have had extensive first aid training. If CERT assistance is required, call security (x-2400); they will dispatch a team.

Pregnancy

If you are, think you may be or are planning to become pregnant, you should be aware that the risks to a mother and a developing foetus from the chemicals used in these laboratories are generally unknown. You may wish to consider deferring this course until after your baby has been born. If you decide to take this course, consultation with your family doctor is strongly encouraged. If you would like the Material Safety Data Sheets for the chemicals in this course, they will be provided.

Fainting

If at any time you feel light-headed, dizzy or that you may faint, immediately sit down on the floor. Fainting itself is rarely harmful; falling because of it may cause injury. Call for medical assistance. If someone should faint, try to assess whether they have been injured by the fall. Make them as comfortable as possible and call for medical assistance.

Cuts

Minor cuts should be treated with plenty of cool water. Ensure no foreign objects are present (such as glass) and apply an appropriate dressing. Seek medical attention at student services.

For more serious cuts the victim should sit or lie down and keep the cut elevated. Bleeding should be kept to a minimum by applying direct pressure (assuming no foreign objects are present in the wound). Call for emergency medical assistance.

Burns

Burns can be either thermal (caused by heat) or chemical. In both cases the first step is to apply plenty of cool water. If the chemical reacts with water, remove it by brushing it from the skin. If necessary, seek medical attention.

Allergies and Other Pre-existing Conditions

If you suffer from severe allergies or other pre-existing medical conditions such as epilepsy, diabetes, etc., it may be helpful (although not required) to alert your instructor(s) and to advise them of any necessary precautions or first-aid treatment.

GOOD LABORATORY PRACTICE

1. Use clean equipment. Dirty apparatus can lead to poor results (and poor grades!) or to unexpected and potentially dangerous reactions. Beakers and flasks can be cleaned with soap and water followed by thorough rinsing with tap water and deionized water. Soap should be avoided when cleaning volumetric glassware such as pipettes, burettes and volumetric flasks because it can be difficult to remove completely.

2. Once a chemical or reagent has been removed from its original container it must **NEVER** be returned to the container. To do so risks contaminating the entire stock and thereby ruining the experiments of others. If you have excess, it is cheaper to throw it away (appropriately) than to risk contaminating the stock.

3. Chemicals are expensive and solutions can be time consuming to prepare. **DO NOT WASTE CHEMICALS.** Read the manual very carefully and take only what you need.

4. Carefully read the labels of reagent bottles to ensure you are getting the right chemicals. Be sure to properly label apparatus containing chemicals or solutions. Accidents may result from the erroneous mixing of chemicals.

5. Remember to re-cap reagent bottles immediately after use. This prevents the waste of chemicals by contamination and spillage. Do not allow caps to become contaminated through contact with the bench top or with other chemicals.

6. Do not remove reagent bottles to your benches. This causes frustration and unnecessary delay for other students.

7. Certain chemicals used in these laboratories are hazardous or present an environmental risk. Dispose of these chemicals in the appropriate waste containers.

8. Set up apparatus so that it is well back from the edge of the bench. All services (gas taps, water taps, electrical outlets, etc.) should be readily accessible.

9. Keep your work area clean, tidy and well-organized. Cluttered work areas can lead to accidents. Clean up all spills quickly. At the end of the laboratory period clean your work area.

10. Do not dispose of solid or insoluble materials in the sinks. Doing so inevitably leads to clogs of the plumbing and to floods.

11. Large spills of acids and bases can be treated with the appropriate spill kit. Afterwards the bench should be washed with plenty of cold water.

12. Mercury spills should be cleaned up immediately. The vapour from mercury is quite toxic. Mercury spills are generally treated with powdered sulphur which reacts with the mercury to form mercury sulphide. The resulting solid is collected and treated as chemical waste.

13. Do not wander aimlessly in the laboratory.

14. Never interfere with the work of other students unless that work presents an immediate hazard to yourself or to others.

15. At the end of the laboratory period clean all equipment and return it to its appropriate place.

16. If solutions or samples must be stored, they must be properly labelled. At a *minimum* the

label must include: i) all the chemical species present and their concentrations; ii) the solvent (when applicable); iii) the date the material was stored and iv) your name. Additional information that should be on the label is: v) the course; vi) the experiment; vii) sample code (when applicable).

Handling Glassware

1. Apparatus that can roll (such as thermometers, etc). should be placed on the bench at right angles to the edge of the bench to prevent it rolling onto the floor.

2. Suction flasks (or Büchner flasks) may collapse violently under vacuum if cracked or otherwise weakened. Inspect suction flasks before using. Do not strike or tap a suction flask while it is under vacuum.

3. Chipped, broken or cracked glassware should be discarded. Heating cracked glassware is very dangerous - the glassware may shatter. Inspect glassware before using.

4. When inserting glass tubing into a stopper match the tubing to the size of the hole. Sometimes the tube can be lubricated with water or glycerol. To protect hands from being cut, wrap tube in a towel before inserting into the stopper. Apply force to the tube length-wise while slowly twisting the tube.

5. To break glass tubing, use a triangular file to scratch the tubing at the point of the break. Moisten the scratch and wrap the tube with a towel. Place thumbs against the glass tubing on the opposite side of the scratch. Press against the tube while pulling hands apart. Fire polish the ends of the tubing before using.

SAFETY ACKNOWLEDGEMENT

Carefully read the following and print and sign your name on the form. You must sign this form and present it to your laboratory instructor at the **beginning** of your first laboratory period.

I have read the safety rules and good laboratory practices outlined at the beginning of this laboratory manual and agree to abide by these rules and practices. I acknowledge that failure to follow these rules may result in dismissal from the laboratory period, a mark of zero for the experiment and no opportunity to repeat the experiment. I accept that persistent failure to abide by the safety rules and good laboratory practices will result in dismissal from the laboratory portion of the course.

I have read the guidelines on laboratory reports. I acknowledge that failure to abide by these instructions may lead to loss of marks.

I have read the instructions on the use of laboratory equipment and agree to use the equipment in accordance with those instructions. I acknowledge that using the equipment in such a way that is dangerous or that is potentially damaging to the equipment may result in dismissal from the laboratory period, a mark of zero for the experiment and no opportunity to repeat the experiment. I accept that persistent abuse of equipment will result in dismissal from the laboratory portion of the course.

Name: _____

Signature: _____

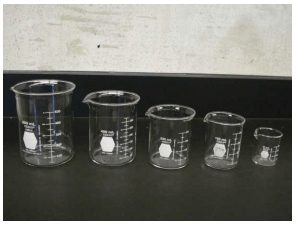
Student Number: _____

Date: _____

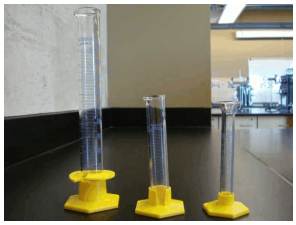
Laboratory Period: _____

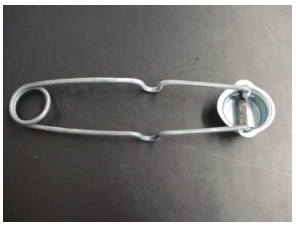
LOCKER EQUIPMENT LIST

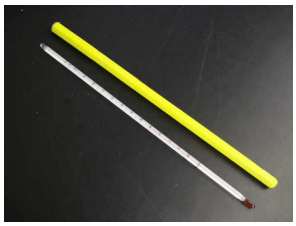
Item	Number
50 mL beaker	2
150 mL beaker	2
250 mL beaker	2
400 mL beaker	1
600 mL beaker	1
50 mL Erlenmeyer flask	1
125 mL Erlenmeyer flask	2
250 mL Erlenmeyer flask	3
10 mL graduated cylinder	1
25 mL graduated cylinder	1
100 mL graduated cylinder	1
Gas lighter	1
Crucible tongs	1
Thermometer, alcohol	1
Tweezers	1
Funnel, short stem, plastic	2
Watchglass, 100 mm	2
Medicine dropper	2
Scoopula	1
Stirring rod	1
Stirring rod with rubber policeman	1
Test tube brush, large	1
Test tube bush, small	1
Test tube rack	1
Test tube holder	1
Test tube, 20 mm x 150 mm	12
Wash bottle, 500 mL	1
Plastic bottle, 500 mL	1


Beakers


Erlenmeyer Flasks

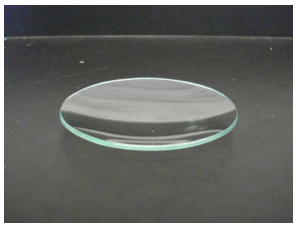

Graduated Cylinders


Gas Lighter


Thermometer

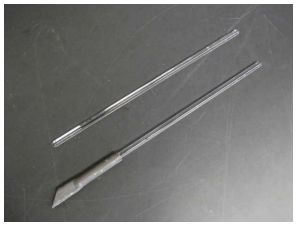

Tweezers

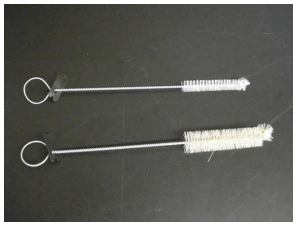

Plastic Funnel

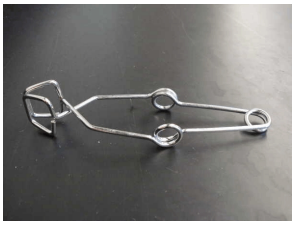

Watchglass

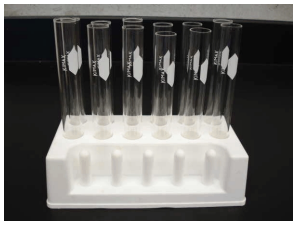

Medicine dropper


Scoopula


Stirring Rods


Test Tube Brushes


Test Tube Holder


20 x 150 mm Test Tubes and Rack


Wash Bottle (l) and Plastic Bottle (r)

Equipment photographs courtesy of Kaitlyn Yarrow

MISSED EXPERIMENTS

For students who miss an experiment (for whatever reason) the first remedy will be to re-schedule the experiment (see below). Where this is not possible (and re-scheduling may be very difficult), a student may be excused from an experiment for "acceptable reasons" after providing appropriate documentation to the senior laboratory instructor. The acceptable reasons for missing an experiment are: illness (or other medical reasons), bereavement, religious observance or court appearance. You may be excused for other reasons at the discretion of the senior laboratory instructor who may require documentation for your absence. Other remedies such as quizzes or assignments in lieu of laboratory reports may also be used (but not necessarily).

In the case of foreseeable circumstances (e.g., varsity athletics, religious observance), requests for re-scheduling and/or being excused from an experiment **MUST** be made at least **seven (7)** working days in advance.

Documentation for missed experiments should be submitted electronically to the senior laboratory instructor. You may either photograph or scan the documentation (include all pages and ensure the documentation is legible) and email it to the senior laboratory instructor: richard.bartholomew@uoit.ca.
If you submit a hard copy of your documentation (or if a hard copy is requested), be sure to make and keep a copy of the documentation for your own records. Any documentation for missed experiments **MUST** be submitted no more than **five (5)** working days after the missed experiment.

If you have missed an experiment for a documented, acceptable reason, any laboratory report that was due at the beginning of the missed laboratory period is due on the first day that you return to campus (the day after the last day given on your documentation). It may be submitted to the senior laboratory instructor.

Re-scheduling Experiments

You are permitted to re-schedule only **ONE (1)** laboratory period without supporting documentation.

You should complete the "Request for a Re-Scheduled Experiment" form available in the "Laboratories" section of the Blackboard site for the course. To complete the form, you must suggest an alternative time to attend a laboratory period.

It is solely your responsibility to find a laboratory period when you can attend. A list of scheduled laboratory periods is available on the "Preview of Available Courses" at MyCampus:
http://www.uoit.ca/mycampus/avail_courses.html
There must be sufficient space in the laboratory period. In other words, the number of students enrolled in the section must be less than the maximum number of students allowed in the section. Note, re-scheduling is done on a first-come, first-served basis; a laboratory period may be full because of re-scheduling even if MyCampus indicates a vacancy.

Once completed, the form (in MS Word format) should be submitted via e-mail to the senior laboratory instructor for the course:
(richard.bartholomew@uoit.ca
for approval. Your request **MUST** be submitted no more than **three (3)** working days after the missed laboratory period and should be submitted at least three days in advance of the laboratory period you wish to attend. You should use the following subject line in your e-mail:

<CHEM XXXX> - request for lab re-schedule, <your name>

If your request is approved, the senior laboratory instructor will sign and return the form to you by e-mail. Approval is solely at the discretion of the senior laboratory instructor and not all requests

will necessarily be approved. If your request is approved, you must present the signed (by the senior laboratory instructor) form to the teaching assistant when you arrive at your re-scheduled laboratory; it is your permission to attend the period. At the end of the period have the teaching assistant sign the form (and any relevant data sheets, laboratory notebooks, etc.). When you submit your laboratory report to your regular teaching assistant, attach the completed form (with all relevant signatures). Failure to do so will result in a grade of zero for the laboratory report. Unless otherwise stated, the report for the re-scheduled experiment will be due at the beginning of your next regularly scheduled laboratory period.

Unless otherwise stated, any laboratory reports that are submitted late as a result of missing a laboratory period will have late days assessed at the usual rate.

LABORATORY REPORTS

Unless instructed otherwise, students will work independently in the laboratory. Attendance at laboratories and submission of a laboratory report are compulsory. Failure to attend a laboratory period or to submit the report will result in a grade of zero for the report for that experiment. Any student who does not submit more than **TWO** (2) laboratory reports will **NOT** receive credit for the laboratory portion of the course. This may result in failure of the course.

1. Laboratory reports are generally due at the beginning of the laboratory period immediately following completion of the experiment unless you are specifically told otherwise. The last report is an exception and is due one week after the last laboratory session. A report that is submitted early will receive 1 day "credit" for each day it is early (not counting week-ends and holidays). A late report will receive 1 day "debit" for each day it is late (not counting week-ends and holidays). At the end of the semester, 2 marks will be deducted from the final laboratory grade for each of the total, accumulated late days. No report will be accepted more than 5 working days late. Reports submitted after 4 pm on Friday will be counted as submitted on the next working day.

2. Once a student has accumulated a total of 5 late days, no other reports will be accepted late (i.e., they will receive a grade of zero) until this "debt" has been reduced. At no time will the debt be allowed to be greater than 5 days. So, for example, if your current debt is -3 days you will not be able to hand in a report more than 2 days late.

3. The laboratory report must be typewritten or written in ink - the choice is yours. Two (2) marks will be deducted if the report is not typewritten or written in ink.

4. White-out (or similar product) must never be used on laboratory reports. Two (2) marks will

be deducted for using white-out (or similar product).

5. The report should include a title page with your name, the date the analysis was completed, the teaching assistant's name, and your laboratory period (day and time).

6. Laboratory reports are not "formal". The reports must include your raw data (with signed copies from your laboratory notebook), sample calculations and graphs, your calculated results and the answers to the questions posed (if any) in the manual.

7. For many of the experiments you will have to analyse unknowns. If an unknown code is provided, you **must** include the code in your report. Failure to do so will result in a grade of zero for the report. Performing an analysis without identifying the sample is useless.

8. Account for all samples and trials. If a result is "rejected", a justification for the rejection must be given. Usually this is a legitimate statistical test and it must be shown to justify the rejection of the data. The percent discrepancy between two values 'a' and 'b' where a < b is given by:

$$\frac{b-a}{a} \times 100\%$$

9. Each analysis should show relevant sample calculations or pre-lab calculations.

10. Where appropriate, clearly state your final result with a 95% confidence interval. If there are replicate determinations, you should include the relative standard deviation of the determinations.

11. Academic misconduct is a serious offence and will be punished. Academic misconduct includes (but is not limited to): plagiarism (copying) of lab reports or spreadsheets,

submitting false data, misrepresenting data or using data from other students without the permission of the instructor. Details about academic misconduct, punishments and appeals procedures are given in the university calendar. It is the student's responsibility to read and understand these regulations.

Marking Schemes

A "performance evaluation" is part of your evaluation in the laboratory and is an assessment by the teaching assistant of your conduct in the laboratory. Preparation, organization, and safety will all be considered.

Teaching assistants are provided with uniform marking schemes for all the experiments. However, within the context of these marking schemes teaching assistants will execute their own judgement. At the end of the semester laboratory grades may be adjusted to account for variation between individual teaching assistants. This may result in an increase or a decrease in your laboratory grade. For semesters in which there is more than one teaching assistant, an overall class average for the laboratory marks will be calculated and the marks for each individual teaching assistant will be scaled to this value. No correction will be applied if the correction would be less than 10% of the final laboratory grade for the course.

The percentage of the final mark of the course assigned to the laboratory will be decided by the professor teaching the course.

Chemistry 2030

Laboratory Reports (Excel Tutorial and Experiments 1, 2, 3, 8)	15%
Determination of Unknowns (Experiments 4, 5, 6, 7, 9)	50%
Pre-lab Questions (Experiments 4, 5, 6, 7, 8, 9)	15%
Laboratory Notebook	10%
Performance Evaluation	10%

Pre-lab Calculations:

For experiments 4, 5, 6, 7 and 9 a set of raw data will be provided. These data should be used to construct a spreadsheet to perform the calculations necessary to determine the quantity (or quantities) of interest in the analysis. The correct result will also be provided so that the calculations can be verified. Submit a print-out of this spreadsheet (along with sample calculations) at the beginning of the laboratory period. If you have trouble with the calculations, you should seek help from an instructor. This spreadsheet can then be used to do calculations with your own data.

In experiments 4, 5, 6, 7 and 9 marks will be assigned on how close your reported value is to the accepted values for the unknowns (the accuracy) and some marks will be assigned for the reproducibility of your results (the precision). The teaching assistant will assume you know how to do the calculations and will assign grades *solely on the basis of the final results you report.* In other words you must be sure that you know how to do the calculations - no consideration of mistakes in calculations will be made in assigning grades. It is crucial that your spreadsheet is correctly designed.

Chemistry 2130

Laboratory Skills and Excel Tutorial	5%
Laboratory Reports for Experiments 1, 5, 7, 8	60%
Laboratory Notebook	10%
Pre-lab Questions (Experiments 5, 7, 8)	15%
Performance Evaluation	10%

Pre-lab Calculations:

For experiments 5 and 7 a set of raw data for the experiment will be posted on the Blackboard site. Use these data to construct your own Excel spreadsheet to calculate the quantity (or quantities) of interest for each experiment. The correct result(s) will also be provided to verify the calculations. At the beginning of the corresponding laboratory submit a print-out of the spreadsheet and sample calculations. The teaching assistant will grade the calculations and provide feedback. When the laboratory report is submitted, include a print-out of the spreadsheet using *your own* data collected in the laboratory. Also include sample calculations using *your own* data.

LABORATORY NOTEBOOK

All students must maintain an up-to-date laboratory notebook. The notebook should be a record of your experiments *as the experiments are performed.* A well-maintained laboratory notebook is a vital aspect of all laboratory experiments. Good laboratory notes allow the re-creation of successful experiments and the determination of why an experiment did not work as anticipated. Good notes are a crucial tool in safety investigations when an experiment goes badly amiss. The laboratory notebook should contain enough detail to allow another chemist to re-create the experiment. It should include the actual steps (as opposed to those prescribed in the laboratory manual) that you took while performing the experiment, any observations that you make (temperature change, colour change, gases evolved, etc.) along with the raw analytical data (masses, volumes, etc.). For example:

> *0.4 g of the unknown were transferred to a weighing vial then weighed on the analytical balance. The solid was transferred to a 250 mL beaker and dissolved in deionized water.*
>
> *When reagent B was added to the analyte a blue precipitate immediately formed with the evolution of a colourless gas.*

The laboratory notebook should not be a simple transcription of the procedure given in the laboratory manual. While that may be useful to you in your preparation, it is not a substitute for recording what you *actually do* in the experiment.

An example laboratory notebook (courtesy of Jennie Eastcott, one of the "Tigers") will be posted on the Blackboard site for the course.

Laboratory Notebook Guidelines

1. The laboratory notebook must be a bound, hardcover notebook. Use only one book. **Do not use a "rough" book.**

2. Label the outside of the book with your name, course name and number, locker number (when applicable), semester, laboratory day and time.

3. Use a pen with indelible ink to record **ALL** entries in the notebook.

4. Write only on the right-hand side of the notebook. The left-hand side may be used for rough calculations, but all data must be recorded on the right-hand pages.

5. All the right hand pages should be numbered sequentially. No pages may be skipped and no pages may be removed.

6. Reserve pages at the front of the laboratory for a table of contents and for a list of the codes for the unknowns (when applicable).

7. Never remove pages from the notebook. Do not erase, remove, or obliterate entries from the notebook. Indicate errors with a single line. The correction should be written nearby. If a page or passage is to be ignored, draw a line through the "offending" passage and give an explanation for the omission. White-out (or any similar material) must **NEVER** be used.

8. Pieces of loose paper must not be used to record data. Data recorded on loose paper will be seized by the instructor.

9. You should note and record any anomalies or difficulties when they are encountered. This

is useful in assessing data when the data are analyzed.

10. The data should be summarized neatly on the right-hand pages.

11. The first page of each experiment should have the following information:
    Date
    Title of Experiment
    References
    Chemical Equations involved in the analysis

    You should prepare this information in advance. You should include the full title, edition and authors of any references.

12. Only data from one experiment should be entered on any given page. For experimental data that must be carried over to another page, label the bottom of the page with "continued on page ..." and on the new page indicate the title of the experiment and "continued from page...".

13. At the end of the laboratory period, have an instructor initial and date each page you have used to record data and observations in that period.

The laboratory notebook will be evaluated to see how well it complies with these guidelines. However, it will not necessarily be collected from every student at the same time. The instructor reserves the right to collect the notebook at anytime and without notice. A mark will be assigned and this will be used to calculate your final laboratory mark.

For chemistry majors: you may use the book for subsequent courses, but do not use one book for more than one course in a semester.

ANALYTICAL CHEMISTRY TECHNIQUES

In all sub-disciplines of chemistry good technique is crucial to obtaining good results and this is particularly true in quantitative aspects of chemistry. To be successful you must master the standard techniques (described below) of analytical chemistry. Links are provided here to videos of the techniques.

Weighing Techniques

Good weighing technique is vital to success in all aspects of chemistry and the measurement of mass is probably the most fundamental measurement in chemistry. Because of advances in technology mass can be one of the most accurately known quantities in any experiment. For greatest efficiency the proper balance must be chosen and the proper procedure must be followed. In chemistry two types of balances (along with specialized variations) are commonly used: the "open-pan" balance and the "analytical" balance.

Open-pan Balance (open-pan balance)

Open-pan balances are used when i) a mass with less accuracy is required; ii) the mass is heavier than can be accommodated on the analytical balance; iii) an approximate mass of a substance must be transferred from one container to another.

Operating an open-pan balance is very simple. Press the "tare" button to "zero" the balance, place the object on the balance pan and record the mass. To measure an approximate mass of substance into a receiving vessel place the receiving vessel on the balance pan and press the "tare" button. This will re-zero the balance. Essentially, the mass of the receiving vessel has been

subtracted. As a substance is added to the receiving vessel, the balance displays the mass of added material. Using an open-pan balance avoids transferring chemicals on the analytical balance.

The Analytical Balance (analytical balance)

The Mettler-Toledo analytical balance used in chemistry laboratories at UOIT is designed to weigh relatively small masses (less than 120 g) with very great precision (to ±0.0001 g). These balances are remarkably easy to use but are very delicate and must be handled with great care.

1. To ensure the greatest accuracy the balance should be level. This can be confirmed by observing the "level indicator" on the balance (which functions exactly like a carpenter's spirit level). The bubble should be exactly in the middle of the circle. If not, consult the teaching assistant.

2. Ensure all the doors are closed. If it is not on already, turn on the balance. The balance will run through a series of checks and eventually should display 0.0000 g. Note: the balance can use different units so make sure the display shows the correct units.

3. Press the "tare" button. This will set the mass reading to zero.

4. Gently slide open one of the doors and place the item to be weighed on the pan. The item must not touch the sides of the balance or the outer ring of the pan.

5. Close all the doors of the balance. Air currents affect the measured mass.

6. Wait until the balance indicates the reading has stabilized and then record the mass.

7. Remove the object and close all the doors.

8. Chemicals must **NEVER** be placed on the balance pan because they may react with it and damage the balance. They must be held in or on some suitable container. Preferably, it is a closed container.

9. If chemicals are spilled in or on the balance, they must be cleaned up immediately. Consult a teaching assistant.

10. Liquids and solutions should generally not be weighed on an analytical balance. If they are weighed on the balance, they **MUST** be in closed containers.

11. Chemicals must not be transferred to a weighing container on (or in) the analytical balance. The transfer should be done using an open-pan balance.

12. Objects which are hot should not be weighed on the analytical balance. The air currents caused by the hot object will cause erroneous readings of the mass.

13. Make sure no one is leaning on the balance table when the measurement is being made as this will adversely affect the measurement.

14. When using the balance for a sequence of measurements on the same object (as, for example, when performing a "weight-by-difference"), it is good practice to use the same balance. This will minimize error because any (constant) error in the balance will be cancelled or partially cancelled in the difference when the difference in mass is calculated.

15. The analytical balance is a very accurate and sensitive instrument. Handling glassware leaves fingerprints and the analytical balance may detect this difference in mass. For greatest accuracy, the weighing vessel should be handled with tongs, while wearing gloves or with a piece of paper towel wrapped around the vial.

16. Substances which are not dry can be difficult to weigh on the analytical balance because the evaporation of water makes it difficult to obtain a stable mass. Similarly, substances which are hygroscopic (water absorbing) are hard to weigh because of the absorption of water.

17. **DO NOT** move the analytical balances.

Weight-by-Difference (weight by difference)

This technique allows very accurate measurement of a relatively small mass transferred to a receiving vessel (e.g., a flask or a beaker). Usually an analytical balance is used, but an open-pan balance may also be used if the masses involved are comparatively large.

1. Using an open-pan balance weigh the (approximate) amount of the substance into a weighing vial. A small beaker may also be used, but a weighing vial is preferable.

2. Weigh the vial on the analytical balance and record the mass.

3. Pour the contents of vial into the receiving vessel.

4. Re-weigh the now (mostly) empty vial on the same analytical balance (see above for the reason why) and record the mass.

The mass transferred to the receiving vessel is simply the difference between the two masses. It does not matter if the weighing vial is completely emptied into the receiving vessel. However, none of the substance may be spilled; all of it must be in either the weighing vial or in the receiving vessel.

Weight-by-difference can also be done in "reverse". That is, the mass of an empty weighing

vial can be recorded. The weighing vial is then filled and the mass of the filled vial is recorded. The mass of the substance in the vial is the difference of the two masses.

An accurate weight-by-difference is best achieved by ensuring the mass of the vial (or beaker) is not too large compared to the mass of the substance in the vial (or beaker). In other words, the difference between the two masses should not be small compared to the two masses.

Volumetric Techniques

Volumetric techniques are probably just as important as weighing technique (in CHEM 2030/2130 volumetric analysis will be used extensively). Good volumetric technique is necessary to obtain good results.

In analytical chemistry there are several important (and accurate) types of volumetric glassware: the pipette (in various forms), the burette and the volumetric flask. Additionally, there are other useful (but not as accurate) pieces of glassware: the graduated cylinder and the Erlenmeyer flask. In the vast majority of analyses, the glassware does not need to be dry to start the analysis. Any residual water (or other solvent) is removed ("exchanged") by systematic rinsing with the solution that will be used in the glassware. This replaces the water with the solution and ensures the solution is not diluted. **NEVER** attempt to "dry" glassware by shaking out excess water. This is an excellent way to break glassware (particularly burettes and pipettes which have delicate tips). Do **NOT** invert glassware (e.g., graduated cylinders, Erlenmeyer flasks, and volumetric flasks) on the bench top in an attempt to drain excess water; glassware in this position is easily knocked over and broken.

Volumetric glassware is normally designated as either "TC" (to contain) or "TD" (to deliver) at an indicated temperature. "TC" means the glassware will contain the designated volume; if the

liquid is drained or poured out, less than this volume will be transferred. "TD", on the other hand, means that if the glassware is drained (or emptied), the designated volume will be transferred to the receiving vessel. The difference between "TC" and "TD" is an important one and should be considered when selecting glassware for an experiment or procedure.

The Meniscus (the meniscus)

To properly use volumetric glassware it is important to understand how to read a "meniscus". When most liquids (including water) are confined to a vessel, they will form a concave surface; the liquid will be higher at the edges than at the centre. This curvature is called the "meniscus" and is caused by the interaction of the molecules of the liquid with the molecules of the vessel. A meniscus is more pronounced for narrower vessels. The meniscus should be read at its lowest point (although in some unusual cases reading the top of the meniscus is required). To avoid parallax error position your eye at the same level as the bottom of the meniscus. The bottom of the meniscus can sometimes be more easily read by placing a white card with a dark stripe behind the glass tube with the top edge of the dark stripe level with the bottom edge of the meniscus. The bottom of the meniscus will be sharper and easier to see.

Transfer Pipette (transfer pipettes)

The transfer pipette is one of the fundamental tools of analytical chemistry. They are accurately calibrated "to deliver" the prescribed volume to the receiving vessel (designated as "TD"). When used properly the error in volume transferred is probably less than 0.2%. The trick is to use it properly! For beginners some practice may be required.

To prepare a pipette for use it should be rinsed with tap water, then with deionized water, and finally with the solution that is to be transferred. The rinsings should be performed three times.

Unless absolutely necessary, do not use soap to clean a pipette. Soap can be difficult to rinse completely from a pipette and may leave a contaminating film.

Squeeze the pipette bulb and place it over the thick end of the pipette (**NEVER** use your mouth!!). Place the thin end (the tip) in the solution. Slowly release your grip on the bulb to draw liquid into the pipette. Fill the pipette about half full. Remove the bulb and quickly place your finger over the end. Hold the pipette nearly horizontal and rotate it so the liquid inside coats the entire inner surface. Drain the rinsings (through the tip) into a waste beaker. Repeat the rinsings twice more. A clean pipette should drain smoothly and leave no droplets on the inner surface of the pipette. If droplets do form, the pipette needs more "vigorous" cleaning. Consult an instructor.

Once the pipette has been properly rinsed a very precise volume can be transferred with the pipette. Use the bulb (**NEVER** use your mouth!!) to draw the liquid into the pipette until the liquid level is well above the graduation mark. For large volume pipettes you may have to squeeze and use the bulb a second (or third!) time. If so, when you remove the bulb quickly place your first finger over the top of the pipette. During the swap you may find it helpful to *gently* rest the bottom of the pipette on the bottom of the beaker. While filling the pipette, do not lift the bottom of the pipette above the level of the liquid. If you do, liquid will squirt into the bulb. This is a very bad thing! It can lead to cross contamination of solutions. Once the level of the liquid is above the graduation mark, remove the bulb and quickly cover the end of the pipette with your first finger. Do **NOT** use your thumb!!

Lift the tip of the pipette out of the solution. Using a Kimwipe or piece of paper towel, wipe excess liquid from the tip. While touching the tip of the pipette to the side of the beaker slowly release the pressure of your finger on the top of the pipette to lower the level of the liquid. Keep the pipette vertical while doing this. Keep your eye level with the graduation mark. Lower the level of the liquid in the pipette until the bottom of the meniscus is exactly at the mark.

To transfer the liquid to the receiving flask vessel, place the tip of pipette against the side of the receiving vessel. Keep the pipette vertical. If necessary, angle the receiving vessel. Remove your finger and allow the pipette to drain. Once the pipette is empty wait ~10 s (to ensure complete and consistent draining) and touch the tip to the side of the receiving flask. Do **NOT** blow out the small amount of liquid in the tip - the pipette is calibrated for the small amount that remains in the tip.

Mohr Pipette (Mohr pipette)

Unlike a transfer pipette (which delivers a fixed volume) a Mohr pipette can be used to deliver variable volumes. The level of accuracy of a Mohr pipette is slightly lower than for a transfer pipette. The technique for using a Mohr pipette is very similar to using a transfer pipette. However, two differences must be kept in mind. First, for a Mohr pipette the volume delivered is determined by the difference between graduations. If the meniscus is lowered from the graduation at 1.00 mL to the graduation at 4.20 mL, the volume delivered to the receiving vessel is 3.20 mL. Second, a Mohr pipette is **NOT** calibrated for the liquid that remains in the tip. If you fill a Mohr pipette and then drain it, you will transfer more solution than you think!

When using a Mohr pipette, it is a good idea to avoid draining to the last graduation on the pipette. If, for some reason, you drain too much from the pipette, you will not be able to determine how much liquid was transferred, so the analysis will be useless.

A Mohr pipette is very useful when multiple transfers (i.e., to different receiving vessels) must be made. As long as the sum of the volumes to be transferred is not greater than the total volume of the Mohr pipette, all of the transfers can be completed with a single fill of the pipette.

Serological Pipette

These types of pipettes are not used extensively in chemistry, but do find wide application in biology and biochemistry. Like a Mohr pipette, they are capable of delivering a variable volume. Unlike a Mohr pipette, they do not operate "by difference". If filled to the 7.00 mL mark, for example, and drained, the pipette delivers 7.00 mL to the receiving vessel. Some serological pipettes require "blow out" of the liquid in the tip; some do not. If you use a serological pipette, you must check to see which type you are using. In this course serological pipettes will not be used.

"Eppendorf"(or micro-) Pipette

Micropipettes are often used in biology and chemistry to transfer small (< 1 mL) volumes. They sometimes have high relative systematic error and variation in volume delivered (from one pipette to another) can be significant. Proper calibration of the pipette is necessary if they are to be used for accurate volumetric work. They must also be re-calibrated periodically as their mechanisms wear over time. Eppendorf pipettes are frequently used improperly leading to significant error. In general, for good (accurate) analytical work a transfer or Mohr pipette is preferred.

Using the pipette is straightforward. Attach an appropriate tip (the tips are often colour coded to the pipette) and depress the button on the top until the "stop" is reached. Hold the pipette vertical and immerse the end of the tip in the solution. Slowly release the button to fill the tip. Place the tip of the pipette against the side of the receiving vessel and push the button all the way to the bottom (past the "stop") to fully discharge the contents. To dispose of the tip press down on the button on the front of the Eppendorf. The pipette should be kept vertical at all times during the transfer. As a general rule: if there is a tip attached, the pipette should be kept vertical. Tipping it on its side can allow the contents of the tip to enter the mechanism of the pipette and damage the mechanism.

Cleaning a Pipette Bulb

If solution is aspirated into the pipette bulb, the bulb must be cleaned to avoid contaminating subsequent solutions. Remove the white taper from the end of the bulb and rinse it thoroughly with deionized water. Rinse the inside of the bulb three times with deionized water and squeeze the bulb repeatedly after each rinsing to expel the water in the bulb. Store the bulb with the open end down to allow any water to drain from the bulb.

Burette / Titration (using a burette, titrating)

A burette is a volumetric device used primarily for titration. It allows variable, but accurately measured, volumes to be added to a receiving vessel. The volume delivered is determined from the difference between a final and an initial volume. A 50 mL burette is typically graduated in 0.1 mL intervals; the volume should be recorded to 0.01 mL by estimating between graduation marks.

A burette is cleaned in a similar way to a pipette. It should be rinsed several times with small portions of tap water, deionized water and finally with the solution that will be used to fill the burette. Before rinsing with the solution, ensure the burette is working properly by draining some deionized water through the tip. Ensure no bubbles are in the tip. Once the tip is bubble-free, ensure the tip is always full. As with the pipette, hold the burette horizontally and rotate it so that the liquid covers the entire inner surface of the burette. Drain the some of the liquid through the stopcock into a waste beaker (ensure the tip remains full). Extra liquid can be poured out the top of the burette. Repeat the rinsings at least twice more.

Using a funnel and a beaker fill the burette with solution to just above the ‘0' graduation. Remove the funnel and open the stopcock and drain some of solution through the burette tip into a waste beaker. Ensure no bubbles are in the tip. Touch the tip of the burette to the side of the waste beaker to remove the last drop. The level of the liquid must be below ‘0'. Record the “initial

volume". When reading the volume, the burette must be vertical and the meniscus should be at eye level. Read the volume at the bottom of the meniscus. You should be able to estimate the volume to 1/10 of a graduation. You may find it useful to use a "burette card" (a white card with a black stripe. The card is placed behind the burette with the top of the black strip at the level of the meniscus) to help read the meniscus. On a burette the volume increases as you read down the burette and indicates the volume that has been *dispensed* from the burette not the volume remaining in the burette.

The burette should be set up with the stopcock facing right (the directions here are for a right handed person; "lefties" should reverse all directions). The stopcock is controlled with the left hand - the palm of the hand is behind the burette and the stopcock is controlled by the thumb and first finger. Initially, this will feel quite awkward but with practice will become more comfortable. The right hand is used to swirl the receiving vessel (normally, a flask). Swirling promotes the mixing of reagents and is crucial to obtaining accurate results. Keep the tip of the burette below the rim of the receiving vessel.

Before beginning the titration, ensure sufficient titrant is in the burette to complete the titration. This is particularly true if you start a titration without having a full burette. If the burette is drained below the last graduation, the volume dispensed cannot be accurately known and the analysis is pointless. Initially, the titrant (the solution in the burette) may be added quickly. As the end-point (the point at which the reaction is judged to be "complete" and the addition of titrant is stopped) is approached, the additions should be slowed down. Very close to the end-point titrant should be added drop-wise (or even ½ drop-wise!). To add drop-wise allow a drop to form slowly on the tip of the burette and then touch the tip to the side of the receiving vessel. Wash the side of the vessel with a little deionized water from a wash bottle. Near the end-point rinse down the sides of the flask to ensure all of the titrant run out of the burette has reacted with the solution in the flask.

Once the end-point is reached wait a few seconds for the burette to drain properly and then

read the final volume. Read the meniscus in exactly the same way as when the initial volume was read.

To clean the burette after the experiment is finished, drain any remaining solution into a waste beaker. Rinse several times with tap water and then with deionized water. Store the burettes upside down (tips facing upwards) and with the stopcocks open.

Graduated Cylinder

A graduated cylinder measures volumes with moderate accuracy; it is better than a beaker, but not as accurate as a pipette. Care should be taken to note whether the cylinder is calibrated "to deliver" (TD) or "to contain" (TC).

Volumetric Flask

A volumetric flask is used to prepare solutions of very accurately known concentration. These solutions can be made by dissolving solids (see Preparation of a Standard Solution) or by diluting other solutions (or liquids). Normally, when the solution is prepared by dilution, the transfer of the solution (or liquid) being diluted is accomplished by using a transfer pipette of appropriate volume (see transfer pipette).

Prior to preparing a solution the volumetric flask should be rinsed several times with deionized water (or the solvent used to prepare the solution).

Once the dissolved solid or solution to be diluted has been transferred to the flask, fill the flask ~3/4 full with solvent and swirl the mixture to ensure good mixing. Continue filling the flask until the meniscus is ~1 cm below the graduation. Complete the filling by adding water drop-wise until the bottom of the meniscus is exactly on the graduation mark. Be *very* careful. If the meniscus

is above the graduation (by even a little bit), you must start again. Once the meniscus is on the graduation, stopper the flask and invert it ("bubble up, bubble down") 20 - 25 times to ensure good mixing. This is crucial step - improper mixing of solutions is a large source of error in undergraduate experiments. While inverting the flask, hold the stopper in place.

Note: it is not necessary to measure (or calculate) the amount of solvent to add to the flask to dilute the solution to volume. Volumes are frequently non-additive (especially when dealing with non-aqueous solvents), so the required volume may be more or less than calculated. Filling the volumetric flask to the mark avoids this problem.

Volumetric flasks are calibrated "to contain" (TC) their designated volume at their designated temperature.

Never put hot (or cold) solutions in a volumetric flask. Never expose the volumetric flask to extremes of temperature. The resulting expansion and / or contraction of the glass may ruin the calibration of the volumetric flask.

Preparation of a Standard Solution (preparing a standard solution**)**

A solution whose concentration is accurately known is a "standard" solution. In many cases a standard solution may be prepared directly from the solid and the solid is referred to as a "primary standard". A primary standard should have the following properties: i) it should be available in high purity (≥99.9%); ii) it should not absorb moisture from the air (i.e., must not be hygroscopic); iii) it should be thermally stable so that it can be dried; iv) it should have high molecular weight to reduce relative weighing errors (because a large mass of the solid must be weighed out to get a given number of moles); v) it should be stable in air. If a substance does not qualify as a primary standard, it can often be "standardized" by titrating it with a solution of a primary standard.

The following is the procedure for preparing a primary standard solution (see, also, instructions for using a volumetric flask).

1. The solid should be dried to constant mass. This ensures the solid contains no moisture and gives a very accurate mass for the solid. Drying to constant mass means drying the solid at > 100^0C, cooling it, weighing it and repeating this process until the mass is constant.

2. Tare an open-pan balance and weigh the required mass of solid into a clean, dry weighing vial. You do not need to record this mass. Make sure the cap is on the vial and re-weigh it on the analytical balance. Record the mass to ±0.0001 g.

3. Pour the solid into a clean beaker of roughly the same volume as the final volume of solution. Do not spill any solid. If you do, you must start again. Do not worry about getting all the solid from the vial into the beaker. It is more important not to spill any.

4. Record the mass (±0.0001 g) of the now (mostly) empty vial using the same analytical balance, again to . The difference between this mass and that recorded in step 2 gives the exact mass of solid transferred to the beaker.

5. Add deionized water to the beaker (approximately ½ the final solution volume).

6. Stir the solution (without spattering) until **all** the solid has dissolved. If you remove the stirring rod at any point, be sure to rinse the stirring rod with deionized water in such a way that the rinsings flow into the beaker. In this way, you will not lose any solid on the stirring rod.

7. You are now ready to begin the quantitative transfer of your solution to the volumetric flask. It is vital that you transfer **all** the solution (therefore, all the solid) to the volumetric flask.

8. Place a funnel in the neck of the volumetric flask. Using the stirring rod as a guide, carefully pour the solution so that it runs down the rod through the funnel into the flask.

9. Using your wash bottle, thoroughly wash the inside wall of the beaker. Transfer the washings to the volumetric flask using the stirring rod and funnel (just as you did in step 8). Repeat the washing and transfer twice more (for a total of 3 times). Be careful not to use too much water in this and all rinsings.

10. Rinse the end of stirring rod using your wash bottle such that the rinsings flow through the funnel into the volumetric flask.

11. Finally, using the wash bottle, rinse the inner surface of the funnel. If steps 8 - 11 have been performed correctly, you will have successfully transferred all the solid to the volumetric flask.

12. Now for the crucial step! Carefully fill the volumetric flask so that the bottom of the meniscus lies exactly on the graduation. It can be neither above nor below the mark. If the meniscus is above, you will have to start again!! The last little bit of water should be added drop-wise using a medicine dropper (or Pasteur pipette) to ensure the flask is not overfilled.

13. After you have filled the flask, invert it 20 - 25 times (while holding the stopper in place) to ensure thorough mixing of the solution.

Suction Filtration (suction filtration)

Suction filtration is an efficient method to separate a precipitate from a solution (the "supernatant"). It can be used when either the precipitate or the supernatant is required. The

technique employs a partial vacuum in a flask to draw the supernatant through a filtering device. Suction filtration is often used to *quantitatively* separate the precipitate from the solution.

Always inspect the suction flask for damage or cracks before use. A damaged flask can implode violently when placed under vacuum and cause serious injury. When using a suction flask, secure it with a ring clamp or an adjustable clamp. Otherwise, it will probably topple over.

For the recovery of small amounts of product, a Hirsch funnel or a sintered glass crucible is used and for larger quantities a Büchner funnel is used. The sintered glass crucible requires no filter paper and is often used when the precipitate must be dried by heating in an oven. For the Hirsch and Büchner filters, a one hole stopper is put on the top of the filter flask and the appropriate funnel inserted into the hole. Sintered glass crucibles use special holders. A thick walled rubber tube connects the side arm of the suction flask to an aspirator which generates the vacuum in the flask.

When using a Büchner Hirsch funnel, place a filter paper sufficiently large to cover all the holes in the funnel and moisten the paper with a small amount of the solvent to ensure the filter paper adheres well to the plate. Turn on the aspirator. A moderate vacuum is created in the flask that will draw the supernatant solution through the filter paper.

To recover the solid, decant (using a stirring rod as a guide) the supernatant solution onto the filter paper and allow it to pass through the filter paper. Next, remove as much as possible of the solid from the beaker onto the filter paper by scraping it out with a "rubber policemen". Small portions of cold solvent can be used to wash out the solid and to rinse off the stirring rod. Do not use too much solvent or the solid may re-dissolve. Get the crystals as dry as possible with the suction filtration. Break the vacuum by lifting the funnel out of the hole (or by removing the tube from the side arm of the flask. Be careful - the side arm is fragile). Wash the solid with the small amount of cold solvent and then re-establish the vacuum. Re-apply the suction until the solid is once again dry. To facilitate better drying of the solid, it can be pressed with the blunt end of a scoopula

to squeeze out any solvent. Large "clumps" of solid can also be broken apart (take care not to tear the filter paper. Repeat these washings 2 - 3 times, breaking the suction before each washing. Several washings with small amounts of solvent are better than one washing with a large amount of solvent. Once the solid is dry, break the suction and *then* turn off the aspirator. It is important to break the suction first. Otherwise, water from the aspirator line may be sucked back into the suction flask thereby contaminating the filtrate. Carefully remove the solid from the filter paper onto a watch glass or into a sample vial. Be careful not to tear the filter paper.

When the supernatant is to be kept, it is a good idea to employ a trap between the aspirator and the suction flask. The trap is a second flask with a two hole stopper and two glass rods in the holes. One glass rod is connected to the aspirator and the other is connected to the side arm of the suction flask. Any water sucked in from the aspirator will be caught in the trap and not contaminate the filtrate. The trap flask must be able to withstand a vacuum - an ordinary Erlenmeyer flask will not do.

Genesys20 Spectrophotometer

The Genesys20 spectrophotometer is remarkably easy to use but is a sensitive and delicate piece of equipment and should be treated accordingly. If, during the course of the experiment, you spill anything on (or in!) the spectrophotometer, immediately clean it up.

1. The lamp in the instrument must warm up prior to use to get the best results. Turn the instrument on roughly 15 minutes before taking measurements. The power switch is at the back of the instrument next to the power cord.

2. The current mode of operations appears in the display. Press the "A/T/C" button to select the desired mode of operation. Unless you are explicitly told otherwise, you will use the

"absorbance" ("A") mode for chemistry experiements.

3. Select the wavelength appropriate for the experiment you are conducting. Press either the nm▲ or the nm▼ key to choose the wavelength. If you hold down the key, the wavelength will change more quickly.

4. Open the sample compartment and insert the cuvette containing the "blank" (a solution containing everything but the species of interest) into the cell holder. Note that the light path is from top to bottom (as opposed to left to right). Make sure the cuvette is positioned so that the light is passing through the clear walls of the cuvette. Do not touch the clear walls of the cuvette as this will leave fingerprints and cause erroneous results. Close the sample compartment door.

5. Press the "0 ABS / 100% T" key to set the blank absorbance to 0 (or, equivalently, % transmittance to 100%). This subtracts any absorbance arising from the blank. Periodically, you should repeat steps 4 and 5 to counter any problem arising from "drift" in instrument response.

6. Remove the blank and insert the cuvette containing the sample into the cell holder. Close the sample compartment door and record the absorbance shown on the LCD display.

7. If you must re-use the cuvette with more than one solution be sure to rinse it thoroughly three times with small portions of the new solution. Start with the most dilute solution and work toward the most concentrated.

Seven-Easy pH Meter

1. Turn the pH meter on.

2. A manual temperature correction must be used to obtain the correct pH. Record the temperature using the thermometer in your locker. Press the thermometer button and use the up and down arrows until the display reads the correct temperature.

3. The electrode should be left in pH 4 buffer when not in use. To ready the electrode for use, lower the rubber safe-lock device (located near the top of the electrode) 2 cm to expose the refilling port (a little hole near the top of the electrode).

4. A 3 point calibration will normally be performed. To begin, remove the electrode from the buffer. Using a wash bottle, rinse the electrode with de-ionized water. Gently **BLOT** excess water from the electrode using a kimwipe. **Do NOT wipe or rub the electrode.**

5. To calibrate the meter immerse the electrode in pH 7.00 buffer and press the "Cal" button. When the calibration has finished, the pH meter will display pH = 7.00.

6. Remove and rinse the electrode and place it in the second buffer (in this case pH 4.00). Press the "Cal" button. Again, when the calibration is finished, the meter will display the correct value (pH = 4.00). Each time you move the electrode from one solution to another you should rinse the electrode and blot it dry. If you are performing only a two point calibration, go to step 8.

7. Repeat step 6 with the third buffer (pH 10.0). At this point the meter will also display the slope and the offset. The slope should be between 95-105%. The offset should be between ±15 mV. If either of these is outside the limits, repeat steps 4 - 7. If it fails a second time,

inform an instructor.

8. Once again, rinse the electrode with de-ionized water, dry it by blotting with a kimwipe, and place it in the solution to be tested. Press the "Read" button to record the pH. Hold the "Read" button for 2 seconds until the "A" on the right side of the display disappears. **IMPORTANT:** ensure the "A" has disappeared before you begin.

9. At the end of the experiment rinse the electrode thoroughly with deionized water, blot it dry with a kimwipe and return the electrode to the pH 4.00 buffer.

EXCEL 2013 - AN INTRODUCTION

Excel (as well as other spreadsheet programmes) is a powerful computational tool with wide application in all areas of science, in chemistry and in analytical and physical chemistry in particular. Calculations on large quantities of data with Excel are quicker and far easier than on a calculator. Because Excel will re-calculate results "instantly" if one of the input variables is changed, it can be easily used to examine the effect(s) of changing variables. Likewise, if an error has been made entering one of the input variables, the correction can be made simply by entering the new value and all the subsequent calculations will be automatically corrected; much easier than re-doing all the calculations by hand!

In junior courses Excel is used primarily for stoichiometric and titration calculations, for statistical functions (such as AVERAGE and STDEV.S), for graphing experimental data and for linear regression. In upper-year courses, more sophisticated calculations will have to be done. This guide will provide very basic instruction on how to use Excel that will allow you to get started if you are unfamiliar with it. There is much more to learn and whole books have been written on it (e.g. "Excel for Chemists - A Comprehensive Guide", 3rd Edition, E. Joseph Billo, Wiley, 2011). To become truly proficient, you will have to explore, experiment and play with the programme (and use the on-line help!). The introductory material that follows will be far more useful if you read it while using Excel.

Navigation and Orientation

When Excel starts you are presented with a blank "worksheet" consisting of numbered rows (vertically) and labelled (with letters) columns (horizontally). Each intersection of a column and a row is a "cell" which is designated using the column letter and the row number: for example, A1, C4, F7. The individual cells hold data: numbers, mathematical formulae, text, etc.

At the top of the spreadsheet is the "Ribbon". The Ribbon has several tabs ("File", "Home", "Insert", etc.). Clicking on each tab gives a set of related tools or operations. For example, clicking on the "File" tab allows you to save your spreadsheet, open existing spreadsheets, print spreadsheets, etc.

Beneath the Ribbon and to the right is the "formula bar" which shows the formula used to calculate the value in the currently selected cell or, if no formula has been used, the contents of the cell.

An Excel file can be organized into several "worksheets". Normally, the data in a given worksheet are related in some way. The different worksheets have different "tabs" at the bottom of the spreadsheet. By default the tabs are labelled as "sheet 1", "sheet 2", "sheet 3" and so on. Each tab can be given a more descriptive (and more useful) title by right-clicking on the tab, selecting "Rename" and entering the new name. Collectively, the worksheets form a "workbook" and when the file is saved all the worksheets are saved together in a single file or workbook. New worksheets can be added to a workbook by clicking the "Insert Worksheet" button (immediately to the right of the last worksheet tab, designated with a plus sign). Alternatively, you can right-click on a tab and select "Insert". Worksheets can be deleted by right-clicking on the tab and choosing "Delete". Worksheets can be moved (either within the workbook or to another workbook) by right-clicking on the appropriate tab and choosing "Move or Copy...".

Navigating around a worksheet is accomplished either by using the mouse or by using a series of key strokes. You can go to a particular cell using the mouse simply by clicking on the cell or by entering the cell address in the box immediately to the left of the formula bar. You can scroll through the worksheet (vertically or horizontally) by clicking on the arrows in the scroll bars (at the bottom and on the right of the worksheet) or by dragging the scroll bar in the appropriate direction. Navigation with keystrokes is accomplished by:

Arrow Keys	Move left, right, up or down by one cell
Ctrl + arrow key	Moves to the last of the continuously occupied cells in the chosen direction
Enter	Move down one cell
Tab	Move right one cell
Shift + Tab	Move left one cell
Home	Move to the beginning of a row
Ctrl + Home	Move to cell A1
Page Up	Move to the top of the window
Page Down	Move to the bottom of the window
Alt + Page Up	Move to the left of a window
Alt + Page Down	Move to the right of a window
Ctrl + End	Move to the last occupied cell (bottom right hand corner)

Entering and Editing Data in a Worksheet

The data in cells can be either numbers, text, or formulae. As the data are entered into the cell they appear in both the cell and in the formula bar. The entry of data in a cell is completed by either pressing Enter (the cursor will move to the cell immediately below the active cell) or by clicking the ✓ on the formula bar. In the latter case the cursor remains on the active cell. To cancel an entry (and restore the previous entry) click ✗ on the formula bar.

Any data that contain text characters (any character other than the digits 0 - 9, decimal point, or *, +, -, ^, $, %) are recognized by Excel as a "text" entry. By default, text entries are left-aligned in a cell while numbers are right-aligned (alignment of data in cells can be changed by formatting the cells). All data that are prefaced with a single quote are recognized as text by Excel. This is

useful if you want to enter numerals as text.

An equation (called a *formula* in Excel) can be entered in a cell. The cell will display the value calculated by the formula while the formula itself will be shown in the formula bar at the top of the worksheet. Formulae usually make use of values (or contents) in other cells (or ranges of cells) by using *cell references* (e.g. A2 or C1:C8). A huge advantage of using Excel (or other spreadsheets) is that if the value in a cell reference is changed the new value calculated by the formula in the cell is automatically updated. Formulae can contain numbers, cell references, arithmetic operations (addition, subtraction, multiplication, division), parenthesis and Excel's worksheet functions (see below).

Some important points about entering formulae:

- A formula must begin with the equals sign (=).
- The arithmetic operations are: addition (represented with "+"), subtraction (-), multiplication (*), division (/) and exponentiation (^).
- Other functions and operations can be represented using Excel's built-in functions (see below).
- The normal hierarchy of arithmetic operations applies. Parentheses can be used to override the normal order of operations.

Consider the following examples in which the value for 'x' is in cell A1:

Mathematical Function	Excel Formula
$f(x) = 2x + 3$	=2*A1+3
$f(x) = \frac{2x^2 + 4x + 1}{x + 2}$	=(2*A1^2+4*A1+1)/(A1+2)
$f(x) = 4.75 + \log_{10} \frac{1 - x}{1 + x}$	=4.75+log10((1-A1)/(1+A1))

Formulae (or parts of formulae) can be copied from one cell to another by copying the formula from the formula bar.

Pressing F2 allows editing of the data in the current cell. Data can be deleted, copied, etc. from the formula bar.

Excel has over 300 built in worksheet functions. In Excel the worksheet function consists of two parts: the name of the function and the *argument* of the function. The argument is the number (or formula) to which the function is applied. In Excel, the argument is contained within a set of parentheses. Within the set of parentheses the data (or cell references) can be separated by commas (for individual data) or by colons (to indicate a range of cells). In this course the most important functions are for mathematics and statistics (the argument is bolded):

Function	Result
LN(***number)***	Calculates the natural logarithm of a number
LOG10(***number)***	Calculates the base-10 (common) logarithm of a number
EXP(***number)***	Calculates the value of 'e' raised to the power of a number
SQRT(***number***)	Calculates the square root of a number
AVERAGE(***number1, number2,...)*** or AVERAGE(***cell reference 1:cell reference 2***)	Calculates the average value of the numbers given or of the specified range.
STDEV.S(***number1, number2,...)*** or STDEV.S(***cell reference 1:cell reference 2***)	Calculates the sample standard deviation of the numbers given or of the specified range.
T.INV.2T(***probability, degrees of freedom)***	Returns the two-tailed inverse of the Student's t-distribution (see experiment 1, CHEM 2030 / 2130)

Function	Result
F.INV.RT(***probability, degrees of freedom [1], degree of freedom [2]***)	Returns the inverse of the right-tailed F probability distribution (see experiment 1, CHEM 2030/2130)
LINEST(***known_ ys, known_xs, const_logical, stat_logical***)	Returns an array of linear regression parameters (see below and Harris, Quantitative Chemical Analysis)
SLOPE(***known_ys, known_xs)***	Calculates the slope of the regression line y = mx + b
INTERCEPT(***known_ys, known_xs)***	Calculates the intercept of the regresson line y = mx + b

Note: for LN( ), LOG10( ), EXP( ) and SQRT( ), "***number***" can also be a cell reference or a formula. In the latter case Excel will apply the function to the result of the formula.

File Management

The commands needed for managing files are found under the File tab in the Ribbon.

The "Open" command can be used to locate and open existing files (workbooks). The "New" command can be used to create a new workbook (by clicking "Blank workbook"). If you click on "Recent", a list of workbooks (Excel files) that have been worked on recently will appear. Clicking on one of these file names will open the workbook.

A workbook currently being used can be saved by choosing "Save" from the File menu. This will save the workbook with its current name. The same thing can be achieved by pressing Ctrl + S. If you want to change the file name (or the folder in which it is stored), choose "Save As" and choose the new folder and enter the new file name.

Editing a Worksheet

Often the first step in editing a worksheet is selecting cells. Selecting cells (or ranges of cells) is accomplished in one of several ways:

- Click on the cell in one corner of the range and, while pressing the left mouse button drag the cursor to the opposite corner of the selection
- Click on the cell in one corner of the range and hold down the Shift key and click a second cell. All the cells between the row and column of the first cell and the row and column of the second cell will be highlighted.
- A complete row (or column) can be selected by clicking on the number (letter) associated with that row (column).
- If the desired range of cells is bounded by empty cells, the range of cells can be selected by using Ctrl + Shift + arrow key to choose in the appropriate direction.

To choose more than one non-adjacent range, hold down the Ctrl key while choosing each separate range. Once a range of cells has been chosen they will be highlighted and the cells can be copied, deleted, or moved (see *Editing a Worksheet* below).

To insert a new column in a workbook, click a column letter to highlight the entire column. Then, right click on the mouse and choose "Insert". The new column will be placed to the left of the highlighted column. To insert more than one column, highlight more than one column. All the new columns will be inserted to the left of the first highlighted column. Rows can be inserted in exactly the same way by clicking on and highlighting the row. New rows are inserted above the highlighted row. Alternatively, rows and columns and can be inserted by clicking on the "Insert" button on the "Home" tab. This will insert one row (above) or one column (to the left) at a time.

Deleting columns or rows is accomplished by highlighting the columns or rows, right clicking the mouse and choosing "Delete". Columns to the right of the deleted columns are shifted to the left; rows below the deleted rows are shifted up. As with insertion the "Delete" button on the

Home tab can also be used.

Individual cells can be inserted by highlighting the cells, right-clicking on the mouse and choosing "Insert". You will be asked whether the existing cells should be shifted down or to the right. You can also insert columns or rows in this way. Deleting individual cells works in a similar way. Right-click the mouse and choose "Delete". Now you will be asked whether existing cells should be shifted up or to the left. Again, columns or rows can be deleted with this method.

Data in cells, ranges of cells, entire rows or entire columns can be copied to other locations in the worksheet, to different worksheets in the same workbook or even to other workbooks. First, select (highlight) the cell or range to be copied. Next, press Ctrl + C (or select "Copy" in the Clipboard group on the Home tab). A dashed line will appear around the cells being copied. Select the destination and press Ctrl + V (or select "Paste" from the Clipboard group). When performing the Paste operation it is best to simply select the cell that will be in the upper left hand corner of the destination range and then press Ctrl + V. If you wish to *move* data, then instead of using Ctrl + C, use Ctrl + X ("Cut" from the Clipboard group).

When data are pasted into a new range, they are pasted with all the attributes (formatting, formulae, etc.) of the original data. This may not always be desirable (e.g. you may wish to paste the results of a formula as opposed to the formula itself). In such circumstances, choose "Paste Special" from the Clipboard group and choose which attributes you wish to include.

If the data that are copied contain numbers or text, the values are duplicated in the new cell. If, however, the copied cell(s) contain formulae, the formulae will be changed because (unless specified otherwise) Excel uses *relative cell referencing* when formulae are copied. This concept is probably best illustrated with an example. Suppose the formula in cell B1 is:

=2*A1+3

If this formula were copied into cells B2, B3, B4...., the formula would be:

=2*A2+3

=2*A3+3

=2*A4+3

etc.

If it were copied into cell C1, the formula would be:

=2*B1+3

Relative cell referencing allows formulae to give the same results if columns or rows are inserted or deleted or if the cell contents are moved. So, if a column were inserted between columns A and B, the formula in cell C1 would be:

=2*A1+3

If the contents of A1 were moved to C3, the formula in B1 would be changed to:

=2*C3+3.

Sometimes using relative cell references is not desirable; when a formula is copied or moved, you may want a reference in the formula to remain fixed to one particular cell. This is done with *absolute cell referencing.* To use an absolute cell reference preface the column letter and the row number with a dollar sign (e.g. A1). If the formula in cell B1 is:

=4*A1+6

and it is copied to the cells B2, B3, B4..etc., each cell will have the same formula:

=4*A1+6

The referred cell in the formula does not change.

A *mixed reference* is a reference such as $A1 or A$1. In the former case the column reference remains fixed, but the row reference will change. In the latter case, the column reference can change while the row reference will be fixed.

Formatting Worksheets

Column width or row height can be changed by clicking on the Format button in the Cells group of the Home tab of the Ribbon. Select "Column Width" or "Row Height" and enter the value for the width or height. Alternatively (and probably easier), position the cursor over the separator bar between two columns (on the right-hand side of the column whose width you want to change). The cursor will change to a double-headed arrow: ⬅|➡. Click the left mouse button and drag to the right or left to increase or decrease the width. An analogous method can be used to change row heights.

In order to make worksheets more readable it is sometimes desirable to "Hide" columns (or rows). Click on the column label to highlight the column, right-click on the mouse and choose Hide. The column will no longer be visible, but all the data and any cell references will be preserved. To reveal a hidden column, highlight the columns on either side of the hidden column, right-click on the mouse an choose "Unhide". In a similar fashion, rows can be hidden and revealed.

Formatting of cells is accomplished by selecting the desired cells then right-clicking on the mouse and choosing "Format Cells...". Several tabs will then be presented:

Number: sets the format for numbers in the cell. This can be scientific notation, number of decimal places etc.

Alignment: sets position of the data within the cell. "Wrap Text" ensures that all text in a cell is visible within the width of the cell (the height of the cell will change as necessary). "Merge Cells" will combine all the selected cells into a single cell.

Font: Allows you to set the font and font attributes (type, size, colour, bold, etc.).

Border: Allows you to set up the borders around the cell.

Fill: Allows you to set the colour of the background (not the data) in the cell.

Printing

Printing functions are found under the "File" tab in the Ribbon. You can choose to print a selected worksheet, a selection of a worksheet or the entire workbook. You can also choose the paper size and whether the print-out will be scaled (e.g., to allow it to fit on a single page). Worksheets (and graphs) can be printed either in portrait or on landscape orientation.

Graphs (or Charts)

Graphs (or charts as Excel calls them) are one of the more important functions in Excel for chemists[1]. Excel offers 11 standard chart types, most of which are of little or no use to chemists; most chart types are useful for displaying financial information (and probably why they are called "charts" as opposed to "graphs"). For chemistry students the most important chart type is the XY (Scatter).

The first step in creating a chart is to select the data (a column of x data and a column of y data) that will be plotted. You may have more than one set of y data for a given set of x data. Each set of y data is a "series". If the x data and y data are not adjacent, hold down the Ctrl key while selecting the separate ranges.

Once the data are selected, click on the "Insert" tab of the Ribbon and choose " scatter (X, Y) or bubble chart" from the "Charts" group. There are five types of scatter plot. Generally, the type to choose is "Scatter" (the names of the types will appear if the cursor hovers over each icon). When you choose one of the types, the chart will be created immediately as an "embedded chart" in the

[1]*Using Excel to create graphs for courses in chemistry is perfectly acceptable, but SigmaPlot may be a better option, offering greater flexibility and wider regression options.*

active worksheet.

Alternatively, the graph may be created by choosing (ensure no data are selected or highlighted) Insert "scatter (X, Y) or bubble chart" and choosing "scatter" (without markers). Click "select data" and "Add" under "Legend Entries (Series)." You can give the series a name and then choose the X values. Next, choose the corresponding Y values. Click "OK".

The chart will appear with a light grey border around it to indicate the chart is "active". Three "Chart Tools" tabs will appear in the Ribbon at the top of the worksheet. These tabs are "Design", "Layout" and "Format".

The "Design" tab allows you change the general appearance of the chart, to change the data that are used for the plot and to move the chart to its own sheet (or to another worksheet). All graphs in chemistry must have the following basic elements: a title and axis labels. This corresponds to "layout 1" in Excel (the use of a legend is optional and can be deleted after the graph is created). To change the layout of an existing chart, click on the chart to "activate" it (the light grey border will form around it and the Chart Tools tabs will appear in the Ribbon), click the Design tab and make the changes that you want. The chart will be automatically changed.

The "Layout" tab allows you to change the appearance of the graph in more detail. For example, you can change the scales on the axes, change the appearance of the axes (e.g. axis title position, tick-marks, gridlines, etc.), change the appearance of the plot area (e.g., borders, colour, etc.), add a trendline and add error bars. The title of the graph can be changed by clicking on the title of the graph (a solid outline box will form around the title) and typing the new title in the formula bar. Likewise, the axis titles can be changed by clicking on the axis title and typing the new title in the formula bar. Changes to the axis format can be made by clicking on "Axes" and choosing either the vertical or the horizontal axis and then choosing "More Axis Options". This will allow you to change various aspects of the appearance of the axes. Under "Axis Options" you can set the

maximum and minimum values for the values of the axis (i.e. you can set the scale of the axis). You can also set the interval between tick-marks (major and minor). In creating a graph, it is important that the horizontal ('x') and vertical ('y') axis scales are chosen in such a way that the plotted data fill the available space. Excel's default choices for the scales of the axes will not necessarily ensure this is the case. The "Layout" tab also allows you to add a "trendline" which is useful if the experiment requires a regression analysis. The trendline option will allow you to add the function of best fit (along with the equation and regression coefficient, if desired) to the graph.

The "Format" tab contains "Shape" and "WordArt" options. The most useful option in this tab is the "Size" group which (as the name implies!) allows you to change the size of the chart. The size of the graph can also be changed by clicking on the chart (to activate it), positioning the cursor over one of the sets of three dots in the chart border and clicking and dragging the cursor. If a set of dots in the corner of the chart border is chosen, the chart is enlarged in both the horizontal and vertical directions such that the ratio of height to width (the aspect ratio) remains constant.

LINEST and Linear Regression

The objective of a linear regression in a scientific analysis is to find the equation of the "line of best fit" to the data. The general equation of a line is:

$$Y = mX + b$$

where 'm' is the slope of the line and 'b' is the intercept of the line. A regression analysis finds the values of 'm' and 'b' of the line that best fits the data.

The LINEST function is the preferred method for performing linear regression in Excel. Harris, Quantitative Chemical Analysis, has a very good example of how to use LINEST for linear regression and that example is the basis for the description that follows.

To perform a linear regression with LINEST, you must first select (or highlight) a 3 row x

2 column block of cells (an array or matrix) in the worksheet. This array will contain the output of the LINEST function. With the 3 x 2 array highlighted enter =LINEST in the formula bar. Excel will first prompt you to choose the y-values. Choose these by clicking and dragging over the y-data (the chosen range of data will appear in the formula bar). Enter a comma and Excel will prompt you for the x-values. As with the y-data, click and drag over the x-data; again, the chosen range of the data will appear in the formula bar. For both the x- and y-data the ranges may be entered directly by entering the cell references in the formula bar. Enter another comma and Excel will ask you if you wish to perform the fit with or without a constant (i.e., you can choose to set b = 0). If you wish to force b to be zero, then enter "FALSE". Otherwise, enter "TRUE". Enter a final comma and choose whether to output "STATS". Entering "TRUE at this point will cause Excel to print out the slope, intercept, regression coefficient and standard errors in the slope and intercept (in general, you will want to have these data). To get the results of the linear regression (the line of best fit), press Ctrl+Shift+Enter (simultaneously). The output from LINEST will be given in the 3x2 array you previously highlighted as:

slope (m)	*intercept (b)*
standard error of slope (s_m)	*standard error of intercept (s_b)*
square of the regression coefficient (R^2)	*standard deviation of y estimate ($s_{Y.X}$)*

From these results a confidence interval (CI) can be calculated for 'b' and 'm'. The confidence interval is a statement of the range in which the true value is expected to lie (at a given confidence level, commonly 95%). It indicates that there is a 95% (for example) chance the true value lies within the stated interval. The size of the range depends on the confidence level, the standard error and the 't' value. The confidence intervals (CI) on 'b' and 'm' are given by:

$$CI = \pm t_{\alpha,df} \times s_b$$

$$CI = \pm t_{\alpha,df} \times s_m$$

where $t_{\alpha, df}$ is the two-tailed 't' value. It can be found in Excel using the T.INV.2T(α,df) function. 'α' is given by:

$$\alpha = 1 - \frac{CL}{100}$$

where CL is the confidence level (most often 95%). 'df' is the degrees of freedom, given by:

$$df = n - 2$$

where 'n' is the number of measurements (points on the line).

When calculating a "prediction" value of Y_0 for a given value X_0, the prediction interval on Y_0 is given by:

$$Y_0 \pm t_{\alpha,df} \times s_{Y_0}$$

where:

$$s_{Y_0} = s_{Y.X} \sqrt{1 + \frac{1}{n} + \frac{\left(X_0 - \overline{X}\right)^2}{\sum_i^n \left(X_i - \overline{X}\right)^2}}$$

$\overline{X}$ = the average of all the X values in the domain

X_i = the X value of the i^{th} measurement

n = the number of calibration points on the line.

One of the more common uses of the regression line is to calculate an X_0 value from an observed Y_0 value. The confidence interval on X_0 (for $Y=mX + b$, but not for $Y = mX$) is given by:

$$X_0 \pm t_{\alpha,df} \times s_{X_0}$$

where

$$s_{X_0} = \frac{s_{Y.X}}{|m|} \times \sqrt{\frac{1}{k} + \frac{1}{n} + \frac{\left(Y_0 - \overline{Y}\right)^2}{m^2 \sum_i^n \left(X_i - \overline{X}\right)^2}}$$

k = the number of replicate measurements of Y_0 (most commonly this is 1)

n = the number of points on the calibration line

$\overline{Y}$ = the mean value of Y for the points on the calibration line

An alternative (although less desirable) to the LINEST function for linear regression is to use the two Excel functions SLOPE and INTERCEPT. These functions will calculate the slope and intercept of the line of best fit, but will not provide any of the statistics that are provided by LINEST. The syntax for the SLOPE function is:

=SLOPE(***known_ys, known_xs)***

The data ranges can be entered either by clicking and dragging over the cells to select them or by entering the cell references directly in the formula bar. The set of known ys and the set of known xs is separated by a comma. The syntax for the INTERCEPT function is:

=INTERCEPT(***known_ys, known_xs)***

and its use is directly analogous to the SLOPE function.

The foregoing is not meant to be an exhaustive description of Excel. The best way to become proficient using Excel is to practise.

LABORATORY SKILLS AND EXCEL TUTORIAL

Introduction

The first laboratory period will be divided in two parts. The first part will be ~2 hours long and will allow you to practise the laboratory skills described in the front of the laboratory manual. The second part (~1 hour) will be a tutorial on Excel. The laboratory period will be most helpful if you have already read the material at the front of the manual on analytical techniques and on Excel.

Below are series of questions on laboratory technique that you should complete and submit *at the beginning of the first laboratory period.* You should answer the "essay" questions *in your own words* without simply copying from the laboratory manual. Answers copied from the manual will be given a mark of zero.

Pre-lab Questions

True or False?

1. When performing a weight-by-difference measurement, the weighing vessel does not need to be rinsed out to complete the transfer of the weighed material.

2. Objects which are hot may be accurately weighed on an analytical balance.

3. When using a pipette the top, middle or bottom of the meniscus can be read with no loss of accuracy.

4. Using soap to clean volumetric glassware is, generally, not a good idea.

5. Draining a Mohr Pipette completely is the proper way to transfer an accurate volume from the Mohr pipette.

6. Transfer pipettes are calibrated for the small volume remaining in the tip. In other words the small volume in the tip does not need to be "blown out" to transfer the designated volume of the pipette.

7. The volumes on the scale of a burette indicate the volumes dispensed to the receiving flask.

8. If you are right-handed, the stopcock of a burette should be handled with your left hand.

9. To measure a volume of 10.00 mL accurately, the best device to use is a graduated cylinder.

10. When filling a burette, the top of the burette should be below eye-level.

"Essay" Questions

1. You have been asked to accurately weigh ~1.5 g of sodium chloride into a 250 mL beaker. Explain how this is done (without weighing the 250 mL beaker). You have been provided with the NaCl, a scoopula, the 250 mL beaker, a small weighing vial, an open pan balance and an analytical balance. The mass does not have to be exactly 1.5 g, but you need to know the mass in the beaker exactly.

2. What are the major differences between a transfer pipette and a Mohr pipette? Which of the two would you use to transfer a volume of analyte solution to use in a titration? Why?

3. Volumetric glassware is usually designated as either "TC at 25^0C" or "TD at 25^0C". What do "TC" and "TD" stand for and what is the difference?

4. When performing a "weight-by-difference" measurement, it is recommended that the same balance be used for both measurements. Why is this beneficial?

5. Describe how to prepare a transfer pipette for use. Explain how to use a transfer pipette to deliver a fixed volume of a solution to an Erlenmeyer flask.

6. Outline all the steps to prepare a standard solution in a volumetric flask.

7. Describe how to prepare a burette for a titration. Assuming an accurate volume of analyte solution has been transferred to a flask, describe how to perform a titration.

Laboratory Skills Tutorial

As mentioned above, in the first part of this laboratory period you will have roughly 2 hours to practise the techniques described at the front of the laboratory manual. The teaching assistant will circulate in the laboratory and provide instruction, hints, etc. At the end of the protocol is a check-list of skills you should practise (and hopefully master!) during the laboratory period. Once you feel comfortable with a technique you should have the TA observe your technique and initial the item on the check-list (the TA may question you about your technique and how you accomplished the task). At the end of the laboratory period you should submit the completed check-list. Marks are assigned for completing the tasks and having the completion signed by the teaching assistant.

Excel Tutorial

After the class has completed the Laboratory Skills Tutorial there will be a tutorial on the use of Excel. The answers to the Excel tutorial questions may be submitted before the end of the laboratory period or at the start of the next laboratory period if you do not have sufficient time to complete them in the laboratory period. If you feel your proficiency in Excel is sufficient for this course, you do not have to stay for the tutorial, but you must complete and submit the exercises.

Excel Tutorial Questions

1. A file of first-year laboratory marks will be posted on Blackboard. A student's final laboratory grade is calculated by taking the average of their six laboratory reports, adding the performance evaluation and dividing the total by 2 to get a grade out of 15:

$$final\ grade = \frac{\overline{laboratory\ reports} + perf.\ eval.}{2}$$

   a. For each student calculate the average mark and standard deviation for the laboratory reports.
   b. Calculate the final laboratory grade for each student.
   c. Calculate the overall class average and standard deviation for the final grade.
   d. Calculate the class average and standard deviation for each laboratory report.
   e. Repeat calculations a - c but excluding experiment 3.

2. Titration with silver nitrate is a classic method of aqueous chloride analysis. The Ag^+ and Cl^- react to form insoluble AgCl:

$$Ag^+(aq) + Cl^-(aq) \rightarrow AgCl(s)$$

   The first step in the analysis is to determine the concentration of the silver nitrate solution

by titrating it with a solution of NaCl whose concentration is accurately known (in this experiment, the NaCl is a *primary standard* solution). Once the concentration of the $AgNO_3$ is known it can be used to find the mass of chloride in the sample. Below are some experimental data from the summer 2012 semester.

a. Calculate the concentration of the NaCl solution.

b. From the titration data for the standardization of silver nitrate calculate the average concentration of the silver nitrate solution.

c. From the titration data of the unknown chloride solution, calculate the average concentration of Cl^- in the 25.00 mL aliquot of the unknown solution (which is the same concentration as in the 250.0 mL of unknown solution).

d. Calculate the mass of Cl^- in the sample of unknown.

e. Calculate the weight percent Cl^- in the sample:

$$wt\%(Cl^-) = \frac{m_{Cl^-}}{m_{tot}} \times 100\%$$

Preparation of Standard NaCl Solution

Weight of NaCl and Vial / g	25.0581
Weight of Vial / g	24.5523
Total Volume of Solution / L	0.2500

Standardization of the Silver Nitrate Solution

Volume of NaCl Used / mL	25.00			
Titration	I	II	III	IV
Initial Volume of $AgNO_3$ / mL	0.12	0.08	0.10	1.12
Final Volume of $AgNO_3$ / mL	22.80	22.74	22.78	23.80

Titration of the Unknown Chloride Solution

Weight of Unknown and Vial / g	25.4983			
Weight of Vial / g	24.8234			
Final Volume of Solution / mL	250.0			
Titration Volume of Unknown Solution / mL	25.00			
Titration	I	II	III	IV
Initial Volume of $AgNO_3$ / mL	0.10	0.04	0.06	0.10
Final Volume of $AgNO_3$ / mL	27.22	27.14	27.16	27.22

3. Brass is an alloy of copper and zinc, typically in a ratio of ~2:1 by mass. The amount of zinc in a brass sample can be measured by atomic spectrometry because there is a linear relationship between the concentration of the zinc in a solution and the amount of light absorbed (the "absorbance", A):

$$A = m\left[Zn^{2+}\right] + b$$

In an experiment to determine the mass percent of zinc in brass, 0.9982 g of brass were dissolved in concentrated nitric acid. The resulting solution was filtered and quantitatively transferred to a 500.0 mL volumetric flask and diluted to the mark (solution A). From Solution A 5.00 mL were transferred to a 250.0 mL volumetric flask and diluted to the mark (solution B). Four 10.00 mL aliquots of solution B were withdrawn and transferred to separate 250.0 mL volumetric flasks and each solution diluted to the mark to yield solutions C - F. The absorbances of these solutions were measured:

Solution	Absorbance
C	0.3318
D	0.3385
E	0.3467
F	0.3530

To calibrate the instrument (i.e., find the relationship between absorbance and concentration) a series of solutions of known concentration of zinc was prepared. To prepare these solutions a standard solution of zinc (25 mg L^{-1}) was used and the following volumes of the standard solution were transferred to 100.0 mL volumetric flasks and the solutions diluted to the mark. The absorbances of these standard solutions were:

Standard	Volume / mL	Absorbance
1	0.60	0.1153
2	1.00	0.1895
3	1.55	0.3002
4	2.00	0.3640
5	2.50	0.4403

a. Calculate the concentrations of zinc in the standard solutions 1 - 5 (for all concentrations, you should use the units mg L^{-1}).

b. From the data for the standard solutions, create a plot of Absorbance (y-axis) versus $[Zn^{2+}]$ (x-axis). Be sure to i) include a proper title for the plot; ii) ensure the data fill the plot area (i.e., choose appropriate scales for the axes); iii) label the axes appropriately.

c. Perform a linear regression (using LINEST) on the data for the standards to

determine the values of 'm' (the slope) and 'b' (the intercept) in the equation above. Include the equation of the line of best fit on the graph and report the correlation coefficient (the r^2 value).

d. Using the equation of the line of best fit from 'c', calculate the concentration of Zn^{2+} in each of solutions C - F.

e. Using the average concentration of Zn^{2+} from solutions C - F, calculate the concentration of solution B. You will need to use the "dilution formula".

f. Calculate $[Zn^{2+}]$ is solution A. You will need to use the "dilution formula" again.

g. Calculate the weight percent Zn in the original brass solution.

(The data were collected by Stephanie Mavilla and Jennie Eastcott - two of the "Tigers" - in October 2006).

Laboratory Skills Check-list

- Preparation of 250.0 mL of standard solution of 0.1 mol L^{-1} NaCl.

- Transfer of 10.00 mL of water using a transfer pipette.

- Transfer of three replicate volumes of ~3 mL (to three separate flasks) using a Mohr pipette.

- Preparation of a burette for titration and proper dispensing of solution from the burette.

- Proper reading of the burette scale.

1. ANALYSIS OF LABORATORY TECHNIQUE

Objectives

1) To practise using some common laboratory volumetric glassware; 2) to practise working with statistics.

Introduction

Good technique is the key to success in analytical chemistry. If technique is poor, errors will be introduced to the analysis and the final result may be very inaccurate. At the very least the precision of the result will be so poor the result will be considered completely unreliable. The ability to accurately and reproducibly measure volumes is crucial when performing titrations and dilutions and this course heavily emphasizes such volumetric analyses.

In this experiment you will examine accuracy and precision by using different volumetric glassware to deliver a fixed, known amount of water to a receiving vessel. Accuracy is a measure of how close a quantity is to the "true" or accepted value. Precision is a measure of how reproducible a result is. You will repeat the delivery 5 or 6 times (depending on the glassware) and calculate statistical parameters. The statistical parameters will be used to evaluate which glassware is most accurate and most precise. You will also be able to assess how reproducible your own technique is.

Imagine a glass vessel designed to contain a volume of water. The vessel has some fixed, "true" (but unknown) volume which we attempt to measure. Because of random error (or imperfect technique) we shall not measure the same volume with each measurement. Statistics provides us

with tools for estimating the "true" volume of the vessel from the finite number of measurements that we can make.

The measurements that we can make (x_1, x_2, ... x_n) form a sample from a much larger (an "infinite") number measurements that could be made of the quantity of interest. The "sample mean" is defined as:

$$\bar{x} = \frac{\sum_i x_i}{n} \tag{1.1}$$

where: n = the number of measurements of the quantity

x_i = the individual measurements.

The $\sum$ symbol is used to indicate the sum of all the terms, x_i. The sample mean (or average) is a statistical estimate of the "true" mean (the average of the "infinite" number of measurements), μ, of the quantity of interest (the volume of the vessel).

The sample standard deviation, s_x, is a measure of the distribution (or variability) of the individual measurements (x_1, x_2, ... x_n) about the <u>sample</u> mean:

$$s_x = \sqrt{\frac{\sum_i \left(x_i - \bar{x}\right)^2}{n-1}} \tag{1.2}$$

(note: this is the sum of the squares of the differences, not the square of the sum of the differences).

where x_i = the i^{th} measurement of the variable 'x'

$\bar{x}$ = the average value for the variable 'x'

n = the number of measurements of the variable 'x'; n –1 is the "degrees of freedom".

The sample standard deviation is a statistical estimate of the standard deviation, σ, of the "infinite" number of measurements. The smaller the sample standard deviation is, the smaller the distribution of individual values about the sample mean is. In other words the measurement is more reproducible and the precision is higher. So, a lower sample standard deviation is desirable in a measurement.

The sample standard deviation can be used to calculate a "confidence interval". The confidence interval is an expression that the "true" mean, μ, lies within some range of the sample mean. The confidence interval is given by:

$$\mu = \bar{x} \pm \frac{t_{\alpha,df}\, s_x}{\sqrt{n}} \tag{1.3}$$

where '$t_{\alpha,df}$' is the "Student's t-value" and is taken from tables that are readily available in statistics textbooks. Values of $t_{\alpha,df}$ for use in equation (1.3) can be found using Excel with the T.INV.2T function. The value of '$t_{\alpha,df}$' depends on both the "degrees of freedom" (df) and the "confidence level". A confidence level is a statement of the probability that the true mean lies within the stated interval. So, a 95% confidence interval is a statement that the true mean, μ, has a 95% chance of being within the stated interval about the sample mean, $\bar{x}$. The value of 'α' is given by:

$$\alpha = 1 - \frac{C.L.}{100} \tag{1.4}$$

For a 95% confidence level, α = 0.05. As an example, the $t_{\alpha,df}$ value for a sample with n = 6 measurements and a 95% confidence level is $t_{0.05,5}$ = 2.571 (df = n-1=5).

The statistical tests performed in this experiment can be used to discover systematic error in an experiment. If two quantities are found to be "statistically different", the difference cannot be ascribed simply to random variation (or error) alone. The difference between the quantities must "real" and indicates the existence of systematic error.

The validity of a technique is often checked by comparing a measured value (the sample mean) with a known (accepted) value. A simple re-arrangement of equation (1.3) (where the "known value" = μ) gives:

$$t_{calc} = \frac{\left|\bar{x} - known\ value\right|}{s_x}\sqrt{n} \tag{1.5}$$

If this calculated 't' value is greater than the tabulated 't' value (at the specified confidence level and degrees of freedom), the measured mean is statistically different from the known value (or the true mean, μ). Equivalently, if the known (accepted) value is not within the stated confidence interval (equation (1.3)), the measured and accepted values are considered to be statistically different (at the stated confidence level).

Another common test applied to scientific results is to compare the results from two different methods of measurement ($\overline{x_1}$ and $\overline{x_2}$, the two sample means from the two different methods) to see if they are statistically different. Again, a 't' value is calculated:

$$t_{calc} = \frac{\left|\bar{x}_1 - \bar{x}_2\right|}{s_p}\sqrt{\frac{n_1 n_2}{n_1 + n_2}} \tag{1.6}$$

(n_1 and n_2 are the number of measurements of x_1 and x_2, respectively). If t_{calc} is greater than the tabulated value (for the experimental degrees of freedom and specified confidence level), the two sample means are statistically different. The "pooled" standard deviation, s_p, is given by:

$$s_p = \sqrt{\frac{s_1^2(n_1 - 1) + s_2^2(n_2 - 1)}{n_1 + n_2 - 2}} \tag{1.7}$$

and the degrees of freedom are:

$$degrees\ of\ freedom = n_1 + n_2 - 2 \tag{1.8}$$

However, equations (1.6) and (1.8) are only valid if the sample standard deviations (s_1 and s_2) are

statistically not different. To determine if s_1 and s_2 are statistically different an "F-test" is performed. An F statistic is calculated:

$$F_{calc} = \frac{s_1^2}{s_2^2} \tag{1.9}$$

The larger standard deviation is always put in the numerator (i.e., F_{calc} is always greater than one). If $F_{calc} > F_{table}$, the two sample standard deviations are statistically different (F_{table} can be found using the F.INV.RT function in Excel). If this is the case, then:

$$t_{calc} = \frac{\left|\bar{x}_1 - \bar{x}_2\right|}{\sqrt{s_1^2/n_1 + s_2^2/n_2}} \tag{1.10}$$

and

$$degrees\ of\ freedom = \left\{ \frac{\left(s_1^2/n_1 + s_2^2/n_2\right)^2}{\left(\frac{\left(s_1^2/n_1\right)^2}{n_1 - 1} + \frac{\left(s_2^2/n_2\right)^2}{n_2 - 1}\right)} \right\} \tag{1.11}$$

Scientifically, it is often more desirable to have a result that has a very low standard deviation but is further from the known (accepted) result than it is to have a result with a high standard deviation very close to the known (accepted) result. In the former case the experiment has been performed very reproducibly; the difference (error) can be ascribed to a systematic error that can be corrected or taken into account. When the reproducibility is very low (high standard deviation) it indicates high random error and this is much harder to correct. Statistically "real" differences indicate systematic differences. If the standard deviation is high, it can be difficult to conclude any observed difference is "real".

Procedure

Your accuracy and precision using several types of volumetric glassware will be tested. The glassware is:

1. 10 mL transfer pipette
2. 10 mL Mohr pipette
3. 50 mL burette
4. 10 mL graduated cylinder
5. 100 mL graduated cylinder
6. 50 mL beaker

The last three of these are in your locker kit; the pipettes and the 50 mL burette will be provided. CHEM 2030 students will use a class A burette (they are designated with an 'A' near the top) and will use the same burette throughout the term.

Instructions for proper use of the pipettes (transfer pipette, Mohr pipette) and the burette (burette) are in the "Analytical Techniques" section of the laboratory manual.

Note: At the end of this experiment CHEM 2130 students should prepare their glucose working standard solution (see part I of experiment 7).

1. Obtain ~400 mL of deionized water in a 600 mL beaker. Measure and record the temperature of the water. You will need the temperature of the water to convert masses to volumes (using density).

2. Weigh an empty 150 mL beaker on the analytical balance and record its mass. After this point only handle the beaker with a Kimwipe, with tongs or while wearing a glove. Deposition of skin oils will lead to errors in the mass and to misleading results in the experiment.

3. Using the 10 mL transfer pipette, transfer 10 mL of water to the beaker.

4. Record the mass of beaker + water (use the same balance used in step 3). Calculate the mass of water transferred. Using the density of water at the measured temperature, convert the mass to a volume.

5. Repeat steps 3 and 4, until you have completed a total of five transfers. Once you have finished all the transfers, empty the beaker, remove any markings and place it in the oven to dry completely. Remove a beaker from the oven and allow it to cool.

6. Repeat the experiment with the next four pieces of glassware. Each time you place a beaker in the oven, remove one and allow it to cool. It does not have to be your own beaker.

7. For the burette repeat the experiment using 10 mL, 5 mL and 1 mL aliquots of water. For each aliquot size use a total of 5 transfers.

Results, Calculations and Questions

For all statistical tests, use a 95% confidence level.

1. Prepare a table (you can import it from Excel) that includes: i) the measuring device; ii) the water temperature; iii) the masses delivered by each device ; iv) the volume (calculated from the water density) delivered by each device. The density of water can be found using the NIST website: http://webbook.nist.gov/chemistry/fluid/. Choose "saturations properties - temperature increments" and choose a temperature range that includes your observed temperature.

2. For each set of data (there should be 8 data sets), calculate the average (sample mean) volume of water delivered, the standard deviation and a 95% confidence interval for the mean.

3. Each piece of glassware was supposed to deliver a nominal volume of 10 mL of water. Statistically, which ones succeeded? Explain with the results of a statistical test.

4. Were the volumes delivered by the 10 mL graduated cylinder and the 100 mL graduated cylinder statistically different? Justify with a statistical test.

5. Based on the results for the 10 mL Mohr pipette and the 10 mL transfer pipette, which is more precise? Which is more accurate? Explain your answer.

6. Generally a 10 mL transfer pipette is more precise (when used properly) than a 50 mL burette for delivering a nominal volume of 10 mL. Do your results agree with this statement? If your results do not agree, suggest reasons why not.

7. Considering all the glassware and all your results, which device would you choose to deliver a nominal 10.00 mL aliquot of water? Explain, considering both the precision and accuracy.

8. A term known as "relative standard deviation" is defined as follows:

$$RSD = \frac{s_x}{\bar{x}} \qquad (1.12)$$

For each of the aliquot sizes delivered with the burette calculate the relative standard deviation. Plot this as a function of average volume (i.e., $\overline{\mathrm{V}}$) delivered. Explain the trend (if there is one).

2. STATISTICAL EVALUATION OF INDICATORS

Objectives

1) To practise titrating; 2) to use statistical tests to determine the best indicator for a given analysis.

Introduction

In this experiment hydrochloric acid will be "standardized" by titration using a variety of indicators to find the end point. "Standardization" is accurately determining the concentration of a species and the standardization of acids or bases is one of the most common analytical procedures in an analytical chemistry laboratory. In a standardization an accurately known quantity of a second reagent is added to the species being standardized. The second reagent is often a "primary standard" - one from which a solution of very accurately known concentration can be made directly (i.e., by weighing a known amount and dissolving it in a known amount of solvent). An acid - base standardization is performed by titration - one reagent (the titrant) is added from a burette to the other (the analyte) in a flask. The point at which the two reactants are present in their stoichiometric ratio is referred to as the "equivalence point". The volume required to reach the equivalence point is used to calculate the concentration of the titrant.

The key to a standardization is detecting when the equivalence point has been reached. This is normally done with an "indicator". An indicator changes some physical property when the moles of the two reactants are present in the stoichiometric ratio. The most common property is colour. Once the change is seen the titration is stopped and the volume of titrant added is recorded. This volume is used to calculate the concentration of the analyte. The point at which the titration is stopped is, not surprisingly, called the "end point". The difference between the end point and the

equivalence point is the "indicator error" (or "titration error") and one goal in a titration is to ensure this error is as small as possible. Minimizing the indicator error involves choosing the best indicator for the job. To minimize indicator error in an acid - base titration the indicator must change colour over a pH range that encompasses the pH at the equivalence point. It should have a colour change that is very easy to see and that can be detected reproducibly. For example, phenolphthalein is an ideal indicator for the titration of acetic acid with sodium hydroxide. Phenolphthalein changes colour between pH 8.0 and 9.6 and the pH at the equivalence point for an acetic acid / sodium hydroxide titration is usually ~8.7. The colour change is from colourless to pink which is very easy to see.

Hydrochloric acid will be standardized with tris(hydroxymethyl)aminomethane (often abbreviated as 'tris' or 'tham') a basic primary standard. The balanced net ionic reaction between 'tris' and hydrochloric acid is

$$H_2NC(CH_2OH)_3(aq) + H^+(aq) \rightarrow H_3NC(CH_2OH)_3^+(aq) \qquad (2.1)$$

The equivalence point lies between pH 4.5 and 5. The titration will be performed with four different indicators and at the end of the experiment the results will be "pooled". By comparing the accuracy and precision of results, the best indicator can be chosen for this titration. The indicators bromothymol blue (BB), methyl red (MR), bromocresol green (BG), and methyl orange (MO) will be tested.

The difference between "precision" and "accuracy" is important. Precision is a measure of the reproducibility of a result and can be assessed by the (sample) standard deviation of a set of results. The standard deviation is a measure of the "width" of the distribution of results about the average (sample mean) result; essentially, the scatter (or variability) of the results. The average value is given by:

$$\bar{x} = \frac{\sum_i x_i}{n} \qquad (2.2)$$

where x_i = the value of each individual measure of x

n = the number of measurements.

The sample standard deviation is given by:

$$s_x = \sqrt{\frac{\sum_i \left(x_i - \bar{x}\right)^2}{n-1}} \qquad (2.3)$$

where x_i = the i^{th} reading of the variable 'x'

$\bar{x}$ = the average value for the variable 'x'

n = the number of measurements of the variable 'x'; n –1 is called the "degrees of freedom".

The smaller the sample standard deviation the more precise a result is. Accuracy, on the other hand, is a measure of how close the result is to the "true" value; the closer it is, the more accurate the result. Accuracy is a harder thing to assess because if the true value is unknown, how can you judge how close to it your result is? Generally, the accuracy of techniques (or methods) is tested by measuring a known quantity to see if the method can reproduce the known value. This is known as "validation". It is entirely possible for a result to be quite precise but inaccurate; this just means the same incorrect result can be found reproducibly! This likely indicates the presence of a systematic error in the procedure. In this experiment the concentration of the hydrochloric acid will be provided, so both the accuracy and precision of the results can be assessed.

A "confidence interval" can be calculated from the sample standard deviation. The confidence interval is an expression that the "true" mean, μ, lies within some range of the sample mean. The confidence interval is given by:

$$\mu = \bar{x} \pm \frac{t_{\alpha,df}\, s_x}{\sqrt{n}} \tag{2.4}$$

where '$t_{\alpha,df}$' is the "Student's t-value" and is taken from tables that are readily available in statistics textbooks. The T.INV.2T function in Excel provides values of $t_{\alpha,df}$ for use in equation (2.4). The value of '$t_{\alpha,df}$' depends on both the "degrees of freedom" (df) and the "confidence level". The confidence level is the probability that the true mean lies within the given interval. So, a 95% confidence interval is a statement that the true mean, μ, has a 95% chance of being within the given interval about the sample mean, $\bar{x}$. The value of 'α' is given by:

$$\alpha = 1 - \frac{C.L.}{100} \tag{2.5}$$

For a 95% confidence level, α = 0.05. As an example, for n = 6 measurements and a 95% confidence level $t_{0.05,5}$ = 2.571 (df = n-1=5).

Statistical difference between two quantities is an important concept in this experiment. Two quantities are said to be statistically different if the difference between them is due to more than just random variation (or random error) in the measurements themselves. In other words, the difference is "real" (systematic error) and not due only to random error. If a measurement has a very high standard deviation (i.e., the measurement is not very reproducible; it has high random error), it will be more difficult to conclude that any difference from another value is real.

A simple re-arrangement of equation (2.4) provides a method of determining whether a measured result is statistically different from a "known" value:

$$t_{calc} = \frac{\left|\bar{x} - known\ value\right|}{s_x}\sqrt{n} \tag{2.6}$$

where "known value" = μ. If t_{calc} is larger than the tabulated Student t value (for the degrees of

freedom and confidence level), the measured value is statistically different from the "known" value. In this situation the degrees of freedom is n-1. Alternatively, if the known (accepted) value is not within the stated confidence interval (equation (2.4)), the measured and "known" values are considered to be statistically different (at the stated confidence level).

Often a quantity is measured by multiple methods (in this experiment, the HCl concentration is determined using different indicators). Two methods will likely give different estimates of the "true" mean, μ. However, what must be tested is whether the two estimates are *statistically* different from one another. This can be done by calculating a t value:

$$t_{calc} = \frac{\left|\bar{x}_1 - \bar{x}_2\right|}{s_p}\sqrt{\frac{n_1 n_2}{n_1 + n_2}} \tag{2.7}$$

and comparing it with the tabulated Student t-value ($t_{\alpha,df}$) at a specified confidence level and degrees of freedom given by:

$$degrees\ of\ \ freedom = n_1 + n_2 - 2 \tag{2.8}$$

$= n_1 + n_2 - 2$). If t_{calc} is greater than the tabulated value, the two estimates are statistically different. The "pooled" standard deviation, s_p, is given by:

$$s_p = \sqrt{\frac{s_1^2(n_1 - 1) + s_2^2(n_2 - 1)}{n_1 + n_2 - 2}} \tag{2.9}$$

However, equations (2.7) and (2.8) assume the standard deviations (s_1 and s_2) are not statistically different. This can be tested by applying an "F-test" and calculating an F statistic:

$$F_{calc} = \frac{s_1^2}{s_2^2} \tag{2.10}$$

The larger standard deviation is put in the numerator (i.e., $F_{calc} > 1$). If $F_{calc} > F_{table}$, the two standard deviations are statistically different (F_{table} can be found using the F.INV.RT function in Excel). If

this is the case, the calculated t-value must be found from:

$$t_{calc} = \frac{\left|\bar{x}_1 - \bar{x}_2\right|}{\sqrt{s_1^2/n_1 + s_2^2/n_2}} \tag{2.11}$$

and

$$degrees\ of\ freedom = \left\{ \frac{\left(s_1^2/n_1 + s_2^2/n_2\right)^2}{\left(\frac{\left(s_1^2/n_1\right)^2}{n_1 - 1} + \frac{\left(s_2^2/n_2\right)^2}{n_2 - 1}\right)} \right\} \tag{2.12}$$

Procedure

Prior to performing this experiment, you should review the procedures for weight-by -difference (weight by difference) and titrating (burette / titration).

The teaching assistant will assign you an indicator.

1. Obtain ~250 mL of standardized ~0.1 mol L^{-1} HCl. The exact concentration of the HCl will be provided after the experiment is complete.

2. Rinse a 50 mL burette 3 times with deionized water and then 3 times with the 0.1 mol L^{-1} HCl (use about 10 mL for each rinsing). Tilt and rotate the burette so that the liquid touches the entire length of the walls of the burette. Drain the liquid through the stopcock.

3. Fill the burette and pass some of the HCl through the stopcock to expel any bubbles in the tip. Let the liquid settle for a minute and then record the (initial) volume to the nearest 0.01

mL.

4. For this experiment dried 'tris' will be provided. Ideally, the 'tris' should be dried to constant mass[2].

5. Use the weight-by-difference technique to weigh an accurate mass (between 0.42 g and 0.50 g) of 'tris' into an Erlenmeyer flask (either 125 mL or 250 mL). Be careful not to spill any.

6. Add roughly 20 mL of HCl (from the burette) to a flask and swirl to dissolve the 'tris'. This is the first 20 mL of the titration. Avoid getting solid 'tris' stuck on the sides of the flask. If 'tris' does adhere to the side of the flask, it can be washed down with a little deionized water.

7. Add 2 - 3 drops of the indicator. Titrate to the end point. The colour changes at the end point are:

   BB: blue to green
   MR: yellow to orange
   BG: blue to green
   MO: yellow to orange.

8. Wait a few seconds for the volume to settle. Record the final volume. Calculate the concentration of the HCl. The first titration will probably be quite "rough"; you are likely to overshoot the end point. Based on the concentration of HCl that you calculate from your first titration, determine the mass of 'tris' to use so that each subsequent titration requires 35

[2]*Drying to constant mass is a process by which a solid is heated (at a temperature > 100^{0}C), allowed to cool to room temperature, and then weighed analytically. The cycle of heating, cooling, weighing is repeated until the solid has a constant mass (meaning two successive masses agree to ±0.3 mg). In this way one is assured the solid is dry.*

- 40 mL.

9. Repeat the titration to obtain six measurements of the HCl molarity. Note: the masses of tris will be different in each flask so the titration volume will be different for each flask.

Results, Calculations, and Questions

You should report your results (and those of the class) in tabular form; see the example below.

1. For each titration calculate the concentration of HCl. It should be a fairly simple matter to set up a spreadsheet to do calculations as you go. Remember to show a sample calculation in your report.

2. You should have at least six concentrations. A 'Grubbs' test (at a 95% confidence interval) - see Harris, 9th Edition, p. 80 - can be used to discard any results (of the six) that are thought to be erroneous. If you discard any results, be sure to show your justification. For your titrations using your indicator calculate:
   i) The average (sample mean) concentration of HCl, C_{avg}
   ii) The sample standard deviation of [HCl], s
   iii) A 95% confidence interval for the concentration of HCl.

3. Post your results (in Excel format) in the course discussion board. You should do this before leaving the lab. Be sure to include the name of the indicator that you used and all the *individual* concentrations of HCl (not just the average) you calculated. Also include your standard deviation, s.

4. Once all the results are posted, for each indicator calculate:
   i) the average (sample mean) concentration of HCl, C_{ave}
   ii) the sample standard deviation in the concentration of HCl, s_x
   iii) the 95% confidence interval for the concentration of HCl.

5. For your indicator, is your "personal" confidence interval greater or smaller than the confidence interval for the class result for the same indicator? Explain the result.

6. For each indicator (using all available data) determine if the average value for the concentration of HCl is statistically different from the known value. Show your statistical tests and a sample calculation.

7. Choose the two indicators that have the greatest difference in the average concentration of HCl and determine whether the difference between the indicators is statistically significant.

8. Considering all the data, which indicator(s) give(s) the most <u>precise</u> results? Which indicator(s) give(s) the most <u>accurate</u> results? Which indicator would you choose to perform this standardization in the future? Why?

Individual data

Trial	Mass of 'tris' / g	V (HCl) / mL	[HCl] / mol L^{-1}
1			
2			
3			
4			
5			
6			

Indicator:_____

Average Value:_____

Sample Standard Deviation:_____

Class (Pooled) Data

Indicator	Total Number of Measurements	Number of Students	Average [HCl]	Sample Standard Deviation, s_x
BB				
MR				
BG				
MO				

3. THE IMPORTANCE OF SAMPLING IN CHEMICAL ANALYSIS

Objectives

1) To determine the "weakest link" in a chemical sampling routine.

Introduction

Chemical analyses can often be broken down into three distinct steps: i) sampling (obtaining a representative sample from the bulk system); ii) sample preparation (converting the sample to an analysable form by, for example, digestion, dissolution, dilution, etc.) and iii) measurement (titration, spectrophotometry, chromatography, etc.). Each one of these steps will have an associated error that can be characterised by a standard deviation, σ, or a variance, σ^2. The variance associated with sampling is designated by σ^2_{samp}; the variance associated with preparation is designated by σ^2_{prep} and the variance associated with measurement is designated by σ^2_{meas}. The total variance in an analysis is the sum of each of these individual errors:

$$\sigma^2_{total} = \sigma^2_{samp} + \sigma^2_{prep} + \sigma^2_{meas} \quad (3.1)$$

In any analytical procedure the analyst seeks to minimize the total error because that improves the accuracy and precision of the results. To do this efficiently the step that has the largest associated error must be identified. The step with the largest error is often called the "weakest link" in the analysis and reducing its error will have the greatest effect on reducing the overall error. Reducing error from the other sources will have an effect, but it will be comparatively small.

Unfortunately, measuring the errors associated with each step is not an easy matter. In some

cases the errors associated with a particular step are a combination of smaller components. This is particularly true of σ^2_{meas}. For example, in a spectrophotometric analysis (such as this experiment) the error associated with the measurement can be broken down into errors associated with the source, the detector, and the optics (collectively, the errors in the spectrophotometer: σ^2_{spec}) and a component associated with positioning of the sample within the spectrophotometer (σ^2_{pos}). The total error can now be written as:

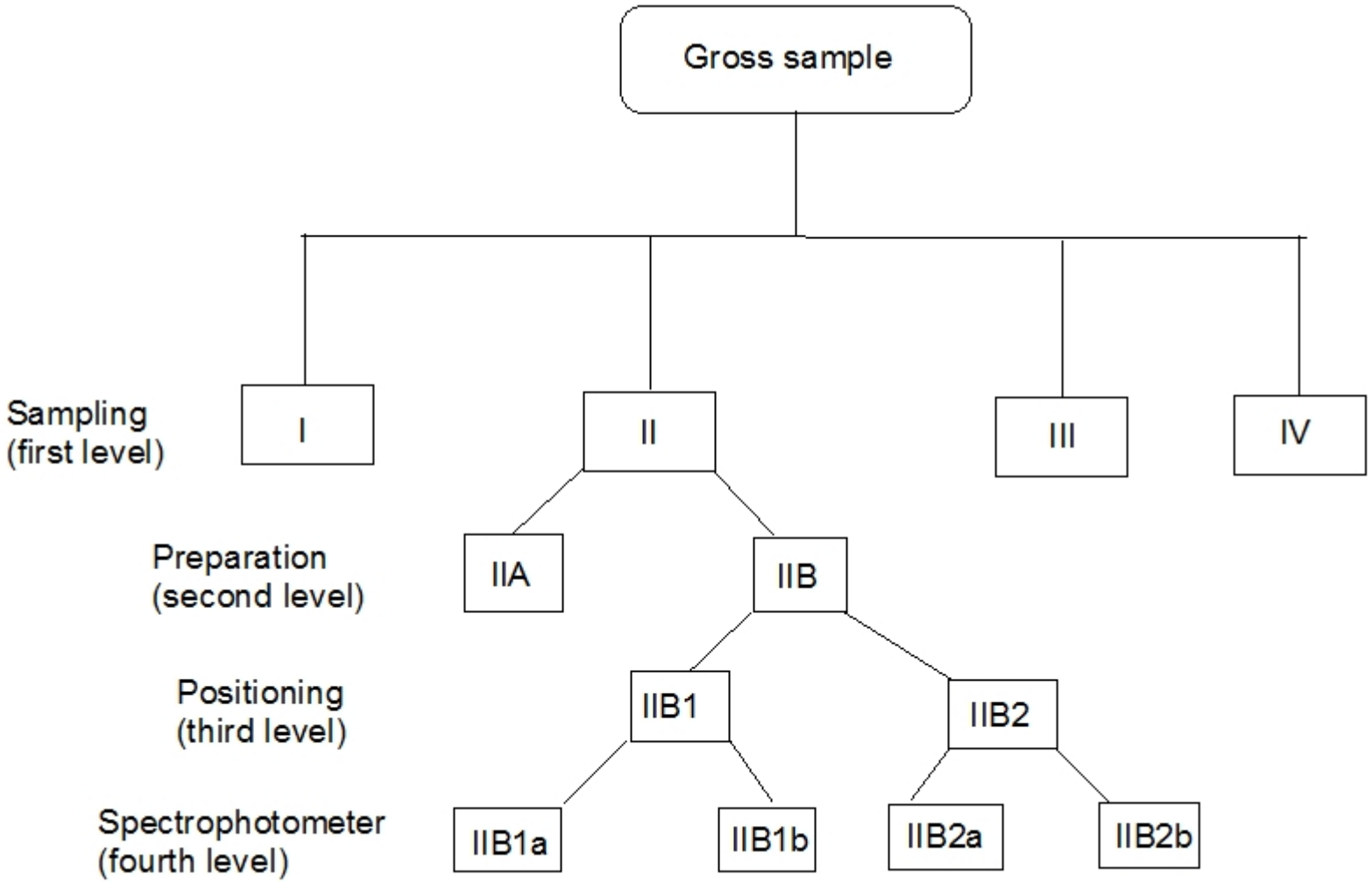
$$\sigma^2_{total} = \sigma^2_{samp} + \sigma^2_{prep} + (\sigma^2_{spec} + \sigma^2_{pos}) \tag{3.2}$$

There are four contributors ("sampling", "preparation", "positioning", and the "spectrophotometer") to the overall (total) error and to extract the individual contributions is non-trivial. One approach is to use an experimental plan known as a "nested design". Such a design consists of several levels and the number of levels equals the number of parameters (the contributors to the error) to be determined. In this experiment the contribution of four sources of error (sampling, preparation, positioning and spectrometer) will be examined, so the experiment requires four levels.

As a matter of statistical principle, the determination of variance requires a very large number ("infinite") of measurements. In a chemistry laboratory this is seldom practical. A much smaller number of measurements is used and the variance, σ^2, is estimated by s^2 - the variance of the smaller number of measurements (the sample variance).

In this experiment four samples collected randomly from the "gross sample" (the original sample being analysed) represent the first level. Each one of these samples is designated with a Roman numeral (I,II,III,IV) and this level will give the error from sampling. Each of these four samples is then ground, divided in two and used to prepare two solutions. This yields eight solutions in total. These 8 solutions constitute the second level and are further designated with the letters A and B (e.g. IA, IB, IIA, IIB, etc.). This second level will give the error associated with preparation.

 UOIT CHEM 2030, S17; Exp. 3 - 12.22



Next, portions of each of these solutions are added to two different cuvettes. This gives the third level (e.g. IA1, IA2, IB1, IB2, etc. - 16 samples) and will yield the error associated with positioning the cuvette (or the cuvette itself). The fourth level of the experiment comes from measuring the absorbance of the solution in a cuvette twice *without removing or re-positioning the cuvette* (IA1a, IA1b, IA2a, IA2b, etc. - 32 measurements). This final level yields the error associated with the spectrophotometer. See page 96.

For the fourth-level measurements, the difference between any two measurements of the same third-level sample (e.g., between IIB1a and IIB1b) is influenced only by the indeterminate errors of the spectrophotometer. The variance of the fourth-level, s^2_{IV}, is an estimate for σ^2_{spec}. However, the variance is determined using differences, so the normal equation for variance is not used. The variance of the spectrophotometer is estimated from:

$$s^2_{IV} = s^2_{spec} = \frac{\sum_i \left(d_{IV}\right)^2_i}{8n} \tag{3.3}$$

where d_{IV} is the difference in weight percent of the analyte (erythrosin B, see below) between *related* fourth-level samples (e.g., between IIB1a and IIB1b, see table below (page 104) - there are 16 values of d_{IV}) and n is the number of first-level samples (in this experiment n = 4). The variance, s^2_{IV}, has 4n degrees of freedom. Note: in equation (3.3) the numerator is the sum of the squares of the differences, d_{IV}, not the square of the sum of the differences.

The third-level variance, s^2_{III}, is influenced by the variance of the spectrometer and the variance of the positioning of the cuvette:

$$s^2_{III} = s^2_{pos} + \frac{s^2_{spec}}{2} = \frac{\sum_i \left(d_{III}\right)^2_i}{4n} \tag{3.4}$$

where d_{III} is the difference between two related third-level average concentrations (e.g., between IIB1

and IIB2, see table - 8 values of d_{III}). The variance, s^2_{III}, has 2n degrees of freedom. The factor of ½ in the s^2_{spec} term accounts for the fact that there are two fourth-level measurements.

The variance for the second-level samples, s^2_{II}, depends on the contributions made from the spectrometer, the positioning of cell and the sample preparation:

$$s_{II}^2 = s_{prep}^2 + \frac{s_{pos}^2}{2} + \frac{s_{spec}^2}{4} = \frac{\sum_i \left(d_{II}\right)_i^2}{2n} \tag{3.5}$$

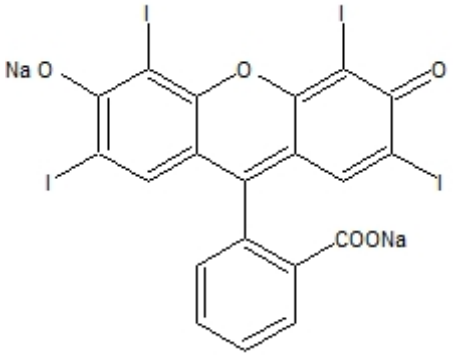
where d_{II} is the difference between related second-level average concentrations (e.g., between IIA and IIB, see table - 4 values of d_{II}). The factors ½ and ¼ account for the two third-level and four fourth- level results used to calculate the second-level result. The second-level variance has n degrees of freedom.

Lastly, the first-level variance, s^2_I, includes contributions from all the sources of error:

$$s_I^2 = s_{samp}^2 + \frac{s_{prep}^2}{2} + \frac{s_{pos}^2}{4} + \frac{s_{spec}^2}{8} = \frac{\sum_i \left(X_i - \bar{X}\right)^2}{n-1} \tag{3.6}$$

where X_i is the result from each first-level sample (I, II, III, IV) and $\overline{X}$ is the average of all first-level concentrations. The variance, s^2_I, has n-1 degrees of freedom.

The analyte in this experiment is erythrosin B:



It is a dye used as a biological stain for bacteria, a plasma stain for nerve cells (in combination with methylene blue) and in quantitative determination of phospholipids. It can be used as an acid-base indicator because it is orange in strongly acidic solutions (pH < 3) and red in solutions at pH > 3. It is also used as a red food colouring. The sample analysed in this experiment is a mixture of erythrosin B (the analyte) and sodium chloride (the matrix). The erythrosin B adheres to the crystals of sodium chloride and makes the solid slightly pink. Because the erythrosin B is coloured and the sodium chloride is not, the concentration of erythrosin B can be determined spectrophotometrically by measuring the absorbance (at a visible wavelength) of a solution of the sample. Basic spectrophotometric analyses depend on the Beer - Lambert Law:

$$A = \varepsilon b C \tag{3.7}$$

The absorbance, A, is linearly dependent on the concentration, C. The molar absorptivity (ε) and the path length (b) are assumed to be constant. Normally, in a spectrophotometric analysis the value of εb would be determined experimentally (through calibration). Here it will be assumed that ε = 0.0916 cm^{-1} L mg^{-1} and b = 1 cm. The concentration of erythrosin B in the <u>solutions</u> can be found from re-arranging the Beer-Lambert Law:

$$C = \frac{A}{\varepsilon b} \tag{3.8}$$

The concentration of erythrosin B in solution can then be used to find the weight percent erythrosin in the *original* <u>solid</u> mixture.

Procedure

This experiment will be performed with a partner. A mixture of erythrosin B and sodium chloride will have been prepared in advance by a technician. This "gross" sample will have been divided into quarters and each quarter placed in a snap cap vial labelled with the Roman numerals I to IV. Each partner should perform the analyses on two of the four "quarters".

Prior to performing this experiment, you should review weight by difference and preparing a standard solution. Review the instructions for the use of the Genesys20 spectrophotometer at the beginning of the laboratory manual (Genesys20 Spectrophotometer). The spectrophotometer will be used at 526 nm.

1. Obtain a roughly 1 g sample from one of the snap cap vials. This is a first-level sample.

2. Transfer the first- level sample to a clean, **glass** mortar. Grind the sample with a **glass** pestle for several minutes to reduce the particle size and to further mix the sample.

3. Divide the ground sample in half and transfer ~0.25 g from one half to a clean weighing vial.

4. Using the weight by difference technique, transfer the ~0.25 g to a clean 50.0 mL beaker. Record the exact mass of the sample transferred. Dissolve the sample in ~20 - 25 mL of deionized water.

5. Quantitatively transfer the solution to a 50.0 mL volumetric flask and dilute with deionized water so the bottom of the meniscus is exactly on the mark. Invert the flask 20 - 25 times to thoroughly mix the solution.

6. Rinse the beaker three times with deionized water to clean it.

7. Repeat steps 3 to 6 with a second 0.25 g portion taken from the glass mortar.

8. Dispose of any solid remaining in the mortar, rinse it with a small amount of deionized water and dry the mortar **thoroughly**.

9. Obtain another 1 g sample from a different snap cap vial of the original gross sample and

repeat steps 2 - 8. Repeat until a sample has been taken from each of the four snap-cap vials.

10. Once the solution preparation is completed, there should be 8 solutions: 2 for each quarter of the original gross sample. These solutions represent the second-level samples.

11. Set the Genesys 20 spectrometer to 526 nm and use distilled water as a blank to "zero" the absorbance.

12. Rinse two cuvettes with deionized water and then with small portions (3 times) of one of the second-level solutions.

13. Fill each cuvette with the first second-level solution. Each cuvette represents a third-level "sample".

14. Insert the first cuvette and record the absorbance. Count to ten and without removing the cuvette record the absorbance a second time. These two absorbances represent a fourth-level "sample".

15. Repeat step 14 with the second cuvette.

16. Repeat steps 12 - 15 with the other second-level solutions.

Once step 16 is complete, 32 absorbances should have been recorded.

Results and Calculations

The calculations for this experiment are very repetitive; the use of a spreadsheet is strongly encouraged. However, you must show a set of sample calculations. The results are best presented in tabular form (see the example given below). In your calculations you should carry more than the normal number of significant figures, but report the final result with the correct number of significant figures.

1. Calculate the concentration of erythrosin B (in mg L^{-1}) in each fourth-level sample using the Beer - Lambert Law (also known as Beer's Law):

$$A = \varepsilon b C \tag{3.9}$$

where A is the measured absorbance, $\varepsilon = 0.0916$ cm^{-1} L mg^{-1}, b is the path length (1 cm) and C is the concentration (in mg L^{-1}).

2. For each fourth-level measurement, calculate the weight percent of erythrosin B in the original gross (solid) sample. Recall that the mass used to prepare the solutions is that recorded in step 4 and that this mass is a mixture of NaCl and erythrosin B.

3. Calculate the weight percent of erythrosin B at each of the third-level measurements (by averaging the weight percents of the corresponding fourth-level values).

4. Calculate the weight percent of erythrosin B at each of the second-level measurements (by averaging the weight percents of the corresponding third-level samples).

5. Calculate the weight percent of erythrosin B at each of the first-level measurements (by averaging the weight percents of the corresponding second-level samples).

6. Calculate the weight percent of erythrosin B in the original, gross sample (by averaging the

weight percents of the corresponding first-level samples).

7. Calculate s^2_{IV}, s^2_{III}, s^2_{II}, s^2_{I} using equations (3.3), (3.4), (3.5) and (3.6). Note: "d_{III}" is the difference between two related third-level average concentrations. Likewise, "d_{II}" is the difference between two related second-level average concentrations.

8. Estimate values for σ^2_{samp}, σ^2_{prep}, σ^2_{pos}, σ^2_{spec} and σ^2_{total} by calculating the corresponding s^2 values.

Questions

1. Are the differences between σ^2_{samp} (3 degrees of freedom), σ^2_{prep} (4 d.f.), σ^2_{pos} (8 d.f.), and σ^2_{spec} (16 d.f.) statistically significant (hint: apply an F test at a 95% confidence limit)? If so, which step is the "weakest link"?

2. How would you improve the overall standard deviation for this analysis?

3. The differences between two fourth-level samples (e.g., between IA1a and IA1b) are influenced only by indeterminate (or random) errors. Explain why. Is this the case for third-level samples (e.g., IA1 and IA2)? What about the second (IA and IB) and first (I and II) levels?

4. The best experimental estimate of the %w/w erythrosin B is the average result of the eight second-level samples. Use the total variance of the analysis to calculate a 95% confidence interval for the %w/w erythrosin B and compare it to the value provided by the instructor. For this calculation there are eight samples and seven degrees of freedom.

sample	A	Mass of sample (from step 5) / g	mg L^{-1} Erythrosin B (from calculaions , step 1)	%w/w fourth level (Step 2)	d_{IV}	%w/w third-level	d_{III}	%w/w second-level	d_{II}	%w/w first-level
IA1a	0.973	0.2642	10.62	0.20103						
					-0.00041	0.20123				
IA1b	0.975	0.2642	10.64	0.20144						
							0.0006198	0.2009238		
IA2a	0.971	0.2642	10.60	0.20061						
					0.00000	0.20061				
IA2b	0.971	0.2642	10.60	0.20061						
									0.03156	0.185
IB1a	0.784	0.2522	8.56	0.16969						
					0.00000	0.16969				
IB1b	0.784	0.2522	8.56	0.16969						
							0.0006493	0.1693612		
IB2a	0.781	0.2522	8.53	0.16904						
					0.00000	0.16904				
IB2b	0.781	0.2522	8.53	0.16904						
IIA1a	0.789	0.2546	8.61	0.16916						
					-0.00043	0.16937				
IIA1b	0.791	0.2546	8.64	0.16959						
							0.001072	0.1688366		
IIA2a	0.785	0.2546	8.57	0.16830						
					0.00000	0.16830				
IIA2b	0.785	0.2546	8.57	0.16830						
									0.01152	0.163

sample	A	Mass of sample (from step 5) / g	mg L^{-1} Erythrosin B (from calculaions , step 1)	%w/w fourth level (Step 2)	d_{IV}	%w/w third-level	d_{III}	%w/w second-level	d_{II}	%w/w first-level
IIB1a	0.735	0.2539	8.02	0.15802						
					-0.00021	0.15812				
IIB1b	0.736	0.2539	8.03	0.15823						
							0.0016124	0.1573166		
IIB2a	0.728	0.2539	7.95	0.15651						
					0.00000	0.15651				
IIB2b	0.728	0.2539	7.95	0.15651						
IIIA1a	0.997	0.256	10.88	0.21258						
					0.00000	0.21258				
IIIA1b	0.997	0.256	10.88	0.21258						
							0.0010661	0.2120505		
IIIA2a	0.992	0.256	10.83	0.21152						
					0.00000	0.21152				
IIIA2b	0.992	0.256	10.83	0.21152						
									0.05641	0.184
IIIB1a	0.724	0.2534	7.90	0.15596						
					0.00000	0.15596				
IIIB1b	0.724	0.2534	7.90	0.15596						
							0.0006462	0.1556345		
IIIB2a	0.721	0.2534	7.87	0.15531						
					0.00000	0.15531				
IIIB2b	0.721	0.2534	7.87	0.15531						

sample	A	Mass of sample (from step 5) / g	mg L^{-1} Erythrosin B (from calculaions , step 1)	%w/w fourth level (Step 2)	d_{IV}	%w/w third-level	d_{III}	%w/w second-level	d_{II}	%w/w first-level
IVA1a	0.782	0.2557	8.54	0.16694						
					0.00000	0.16694				
IVA1b	0.782	0.2557	8.54	0.16694						
							0.0004269	0.1667227		
IVA2a	0.78	0.2557	8.52	0.16651						
					0.00000	0.16651				
IVA2b	0.78	0.2557	8.52	0.16651						
									0.00805	0.163
IVB1a	0.747	0.2557	8.16	0.15946						
					0.00021	0.15936				
IVB1b	0.746	0.2557	8.14	0.15925						
							0.0013876	0.1586641		
IVB2a	0.74	0.2557	8.08	0.15797						
					0.00000	0.15797				
IVB2b	0.74	0.2557	8.08	0.15797						

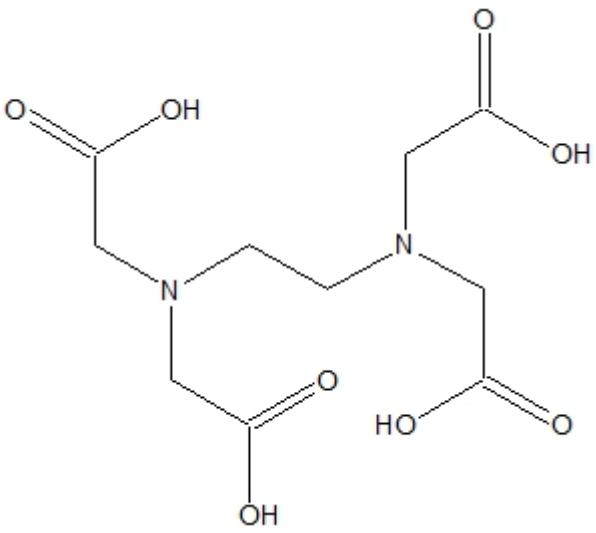
4. THE DETERMINATION OF [Ca^{2+}] BY AN EDTA TITRATION

Objective

1) To measure the total [Ca^{2+}] in an aqueous sample by titration with ethylenediaminetetraacetic acid

Introduction

Ethylenediaminetetraacetic acid (commonly abbreviated as EDTA) forms very stable complexes with a wide range of metal ions in aqueous solution. Because the complexes are so stable, the reaction between the metal ion and the EDTA is essentially quantitative. That is, the equilibrium lies well towards products so that <u>all</u> the metal reacts with the EDTA making EDTA a useful reagent in analytical chemistry.



EDTA

EDTA is a potentially hexadentate ligand. It can bond to a metal ion through six sites: four carboxylic acid groups (-COOH) and two amine groups (-NR_3). In a reaction between a metal ion and EDTA the metal ion acts as a Lewis acid (because it accepts electron pairs from EDTA) and the EDTA acts as a Lewis base (because it donates electron pairs to the metal).

In this experiment the EDTA reacts with the Ca^{2+} ion in aqueous solution in a 1 : 1 mole ratio. Aqueous, fully protonated EDTA has 6 dissociation reactions: 4 for the carboxylic acid groups

and 2 for the (protonated) amino groups. They can be written as:

$$H_6Y^{2+} \rightleftharpoons H^+ + H_5Y^+ \quad (4.1)$$

$$H_5Y^{2+} \rightleftharpoons H^+ + H_4Y \quad (4.2)$$

$$H_4Y \rightleftharpoons H^+ + H_3Y^- \quad (4.3)$$

$$H_3Y^- \rightleftharpoons H^+ + H_2Y^{2-} \quad (4.4)$$

$$H_2Y^{2-} \rightleftharpoons H^+ + HY^{3-} \quad (4.5)$$

$$HY^{3-} \rightleftharpoons H^+ + Y^{4-} \quad (4.6)$$

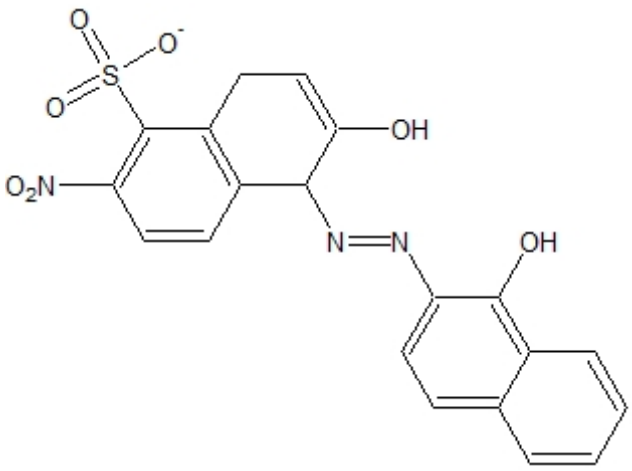
At pH 10 the predominant form of the EDTA is HY^{3-}. The sample will be buffered (using an ammonia / ammonium chloride buffer) at pH = 10 so that two reactions occur:

$$HY^{3-} \rightleftharpoons H^+ + Y^{4-} \quad (4.7)$$

$$Ca^{2+} + Y^{4-} \rightleftharpoons CaY^{2-} \quad (4.8)$$

The equilibrium constant for reaction (4.8) is very large (the reaction essentially goes to completion), consuming all the Y^{4-} produced by reaction (4.7). This, in turn, "pushes" reaction (4.7) towards products (Le Châtelier's Principle). The net effect is that the stoichiometry between the calcium and the EDTA is 1:1.

All the species in this reaction are colourless, so to detect when all the Ca^{2+} has been consumed (the end point) an indicator must be added. The indicator used in this experiment is eriochrome black T which has three acidic protons (in the figure it has lost one proton). At pH 10 the indicator is in the Hin^- form and will form red complexes with metal ions (in this experiment with Ca^{2+} to form CaHIn). This complex is weaker than the EDTA complex so EDTA displaces the indicator and forms its own complex with the



Eriochrome Black T

metal. Before the end point there is a very small amount of Ca^{2+} bound to the indicator and a large amount of "free" Ca^{2+}. The EDTA reacts, initially, with this free Ca^{2+}. Once all the free Ca^{2+} has been consumed (the equivalence point) the EDTA reacts with the Ca^{2+} bound to the indicator, releasing the indicator:

$$CaHIn(aq) + Y^{4-}(aq) \rightarrow CaY^{2-}(aq) + HIn^{2-}(aq) \quad (4.9)$$

The "free" indicator (HIn^{2-}) is blue so the colour change is from red (CaHIn) to blue (HIn^{2-}).

Unfortunately, the formation constant of CaHIn is not very large, so it does not give a sharp end point. This problem is overcome by adding a small amount of Mg^{2+} to the EDTA solution. The formation constant of the magnesium - EDTA complex is smaller than for the calcium - EDTA complex so when the EDTA solution (the titrant) is added to the calcium solution (the analyte) the magnesium is displaced by the calcium:

$$MgY^{2-}(aq) + Ca^{2+}(aq) \rightarrow Mg^{2+}(aq) + CaY^{2-}(aq) \quad (4.10)$$

The displaced magnesium reacts with the indicator:

$$HIn^{2-}(aq)\ \{blue\} + Mg^{2+}(aq) \rightarrow MgHIn(aq)\ \{red\} \quad (4.11)$$

and turns the solution red. Once all the Ca^{2+} has been consumed the first excess of EDTA reacts with the magnesium - indicator complex, releasing the indicator (as HIn^{2-}):

$$MgHIn(aq)\ \{red\} + Y^{4-}(aq) \rightarrow MgY^{2-}(aq) + HIn^{2-}(aq)\ \{blue\} \quad (4.12)$$

which turns the solution blue. This gives the colour change (from red to blue) that marks the end point.

EDTA cannot be prepared as a primary standard. Hence, it will be standardized by titration with a standard solution of Ca^{2+}. The Ca^{2+} solution (as $CaCl_2$) is prepared from calcium carbonate which is available as a primary standard. Calcium carbonate is not soluble in water. To get the Ca^{2+} into solution the $CaCO_3$ is reacted with dilute hydrochloric acid:

$$CaCO_3(s) + 2HCl(aq) \rightarrow CaCl_2(aq) + CO_2(g) + H_2O(l) \quad (4.13)$$

Once the $CaCO_3$ has dissolved the excess HCl is neutralized by adding NaOH, yielding a solution with accurately known Ca^{2+} concentration.

Pre-lab Calculations

1. Using the experimental data in the Excel sheet posted on Blackboard, calculate the concentration of Ca^{2+} in the *original* unknown. Report a relative standard deviation.

Procedure

Prior to performing this experiment you should review preparing a standard solution, using a transfer pipette and performing a titration.

Part I: Preparation of Calcium Chloride Solution

1. Obtain ~ 0.6 g of calcium carbonate ($CaCO_3$) and dry it for one week in an oven at 110^0C. Store and cool the $CaCO_3$ in a dessicator for ~20 minutes.

2. Using the weight-by-difference technique, transfer 0.4 - 0.45 g (accurately known) of $CaCO_3$ to a 250 mL beaker. Add ~100 mL of deionized water. $CaCO_3$ is insoluble, so if the solid dissolves, you have the wrong stuff.

3. Calculate the volume of 1.0 mol L^{-1} HCl required to dissolve the $CaCO_3$ (do not use too much extra HCl). Slowly add this calculated volume of HCl to your $CaCO_3$. After the HCl is added, warm the solution gently on a hot plate. Gas (CO_2) should evolve. Once no further gas evolution occurs, ensure all the $CaCO_3$ has dissolved. If some solid remains, add a little more HCl (2 - 3 drops) followed by gentle warming until all the solid dissolves.

4. Add one or two drops of methyl red indicator (do not agitate the bottles of methyl red). The solution should be bright pink or red. Slowly add 1.0 mol L^{-1} NaOH (with continuous

stirring) to neutralize the excess HCl. The colour will change to pinkish/orange once the solution is neutralized. If too much NaOH is added, the solution turns yellow. If this happens add 1 mol L^{-1} HCl drop-wise to get back to the pinkish/orange colour.

5. Quantitatively transfer this solution to a 500.0 mL volumetric flask and dilute the solution to the graduation mark. Cap the flask and invert 20 - 25 times to mix the solution well. You now have a solution of accurately known Ca^{2+} concentration.

Part II: Standardization of EDTA Solution

1. Collect ~200 mL of EDTA in a clean, dry beaker.

2. Perform a "blank" titration. Put 100 mL of deionized water, 1 mL of buffer and 5 drops of eriochrome black T indicator into a 250 mL Erlenmeyer flask. Titrate with the EDTA solution. The volume should be very small, so titrate with ½ drops. Subtract this "blank" volume from the titration volumes of the other runs. If the solution is blue upon addition of the indicator (i.e., no calcium is present), the blank value is zero. This will allow you to see what the colour at end point should be.

3. Using a properly prepared 25.00 mL pipette, transfer 50.00 mL of the $CaCl_2$ solution to a clean 250 mL Erlenmeyer flask. Add 50 mL of deionized water to the flask.

4. Add 1 mL of ammonia - ammonium chloride buffer to the flask. Add 5 drops of eriochrome black T indicator and titrate with the EDTA solution <u>immediately</u>. The buffer and indicator should only be added immediately before beginning the titration. If the buffer is added too early, $Ca(OH)_2$ may precipitate (as the solution sits). The reaction between calcium and EDTA is "slow", so swirl the flask well and continuously. The colour change at the end point is from wine red to pure blue.

5. Repeat the titration at least two more times using 50.00 mL aliquots of $CaCl_2$ each time.

6. Calculate the concentration of the EDTA solution. It is now standardized and can be used to determine the $[Ca^{2+}]$ in the sample in part III.

Part III: Determination of the $[Ca^{2+}]$.

The procedure for titrating the unknown is the same as for standardizing the EDTA solution in part II. A sample of unknown $[Ca^{2+}]$ will be provided. Note the sample code for your unknown. Report this sample code or you will be given a grade of zero.

1. Collect ~30 mL of the calcium unknown in a clean, dry beaker. The beaker must be dry to avoid diluting the solution.

2. Use a properly prepared pipette to transfer 10.00 mL of the calcium unknown to a 250.0 mL volumetric flask. Dilute to the mark with deionized water. Invert the flask 20 - 25 times.

3. Using a properly rinsed pipette, transfer 25.00 mL of the sample to a clean Erlenmeyer flask. Add 50 mL of water, 1 mL of buffer and 5 drops of indicator solution. Immediately titrate with the standardized EDTA solution to the blue end point.

4. If the titrant volume is below 25 mL, calculate how many extra 25.00 mL aliquots of the sample of unknown $[Ca^{2+}]$ are required to get a titrant volume between 25 mL and 50 mL. Add this volume to the sample in the flask (but do not use a total sample size greater than 75.00 mL) and continue the titration to a blue end point. Use this new, total aliquot size (of calcium unknown) for the remaining titrations (at least 2).

5. If the titrant volume is between 25 and 45 mL, then use a 25.00 mL aliquot for the remaining

titrations (at least 2).

Results and Calculations

1. Calculate and report the total concentration Ca^{2+} in the *original* unknown. Report an average concentration with a relative standard deviation.

5. MEASUREMENT OF VITAMIN C (ASCORBIC ACID) WEIGHT PERCENT BY IODOMETRIC "BACK" TITRATION

Objective:

1) To measure the mass percent of vitamin C (ascorbic acid) in a mixture of solids.

Introduction

In this experiment the amount of vitamin C in a sample will be determined using a "back titration". In a back titration an accurately known excess amount of a standard reagent is added to the analyte. The analyte is completely consumed, but the standard reagent is not. Then, the amount of standard reagent remaining (the excess) is determined by a titration (the back titration). Knowing how much standard reagent was added and the excess allows the calculation of how much reacted with the analyte. Then, the concentration of the analye can be performed.

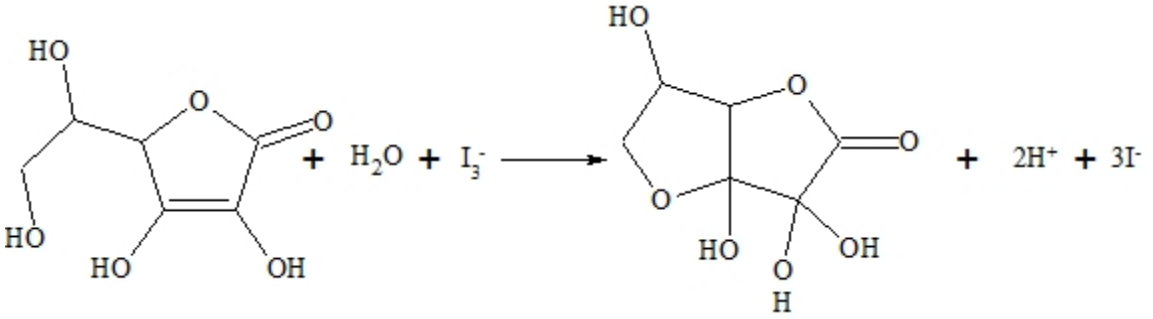
An iodometric titration is one in which the amount of iodine (usually in the form of the tri-iodide ion, I_3^-) is measured by titrating it with a standardized solution of thiosulphate:

$$I_3^-(aq) + 2S_2O_3^{2-}(aq) \rightarrow 3I^-(aq) + S_4O_6^{2-}(aq) \qquad (5.1)$$

Aqueous solutions of the I_3^- ion are, when sufficiently concentrated, deep red / brown. Eventually, as the concentration of I_3^- decreases (through reaction with $S_2O_3^{2-}$), the solution becomes pale yellow. Starch indicator is added at this point. The I_3^- and starch form a complex which makes the solution intensely blue. More $S_2O_3^{2-}$ is added and as soon as all of the I_3^- is consumed the solution changes from blue to colourless. This marks the end point of the titration. The iodometric titration is popular because the end point is very sharp (the colour change happens quickly and is easy to see). However, the starch indicator must not be added too early or some of the iodine remains bound to the starch

after the equivalence point is reached and the colour change at the end point is not clearly observed.

In this experiment the amount of vitamin C (ascorbic acid) in a sample will be determined by adding an excess and well-known amount of I_3^- and then determining the amount of (unreacted) I_3^- that remains. Vitamin C is a mild reducing agent, and, under acidic conditions, will react with I_3^- in a one-to-one ratio:



(5.2)

The remaining (or excess) amount of I_3^- is found by titration using reaction (5.1) (this is the back titration). If the original amount of I_3^- added is known and the excess is measured, the amount of I_3^- that reacted with ascorbic acid can be easily calculated:

$$n_{I_3^-,added} = n_{I_3^-,reacted} + n_{I_3^-,excess} \tag{5.3}$$

From these data the amount of ascorbic acid can be calculated.

The I_3^- can be generated *in situ* by the reaction of KI with KIO_3 and hydrochloric acid. The tri-iodide ion is generated through a disproportionation reaction (a reaction in which one species is both oxidized and reduced):

$$IO_3^-(aq) + 8I^-(aq) + 6H^+(aq) \rightarrow 3I_3^-(aq) + 3H_2O(l) \tag{5.4}$$

KIO_3 is a primary standard, so a solution of it can be made directly from the solid with precise concentration. If the potassium iodate (KIO_3) is the limiting reagent and the precise amount of it used in reaction (5.4) is known, the amount of I_3^- generated is precisely known. An acidified solution of iodate plus iodide can, therefore, be used to standardize thiosulphate solutions (as will be done

in this experiment) according to reaction (5.1). Reaction (5.4) is also used to generate a known amount of I_3^- ($n_{I_3^-,added}$) *in situ* to react with the vitamin C.

This method of analysis is well-suited to determining the concentration of vitamin C in vitamin tablets. The tablet is ground, subdivided and treated with I_3^-. The excess I_3^- is titrated with the thiosulphate. Unfortunately, difficulties can be encountered because of insoluble binding material in the tablets and, more importantly, with sampling errors (c.f. experiment 3). For this experiment the process will be simplified: vitamin C will be provided in a finely ground mixture with a soluble, inert material. The entire mass of the sample will be dissolved, obviating problems with sampling error.

Pre-lab Calculations

1. Using the experimental data in the Excel sheet posted on Blackboard, calculate the weight percent vitamin C in the unknown. Report a relative standard deviation.

Procedure

Prior to performing this experiment you should review weight by difference, preparing a standard solution, using a transfer pipette and performing a titration.

Part I: Preparation and Standardization of Sodium Thiosulphate

1. Retrieve your sample of dried KIO_3 from the oven and place it in a dessicooler. Allow it to cool for roughly 20 minutes.

2. Boil ~500 mL of deionized water for approximately 5 minutes. This helps to expel CO_2 which can cause decomposition of the thiosulphate by lowering the pH of the solution.

3. Allow the water to cool and then add ~0.05 g of Na_2CO_3. This is added to raise the pH and prevent acid induced decomposition of the thiosulphate.

4. Dissolve ~8.7 g of $Na_2S_2O_3 \bullet 5H_2O$ in the 500 mL of water. Aqueous sodium thiosulphate is both air and light sensitive. If it must be stored for a long time, put it in a tightly stoppered, amber bottle or in a cupboard.

5. Once the KIO_3 has cooled, dissolve ~1 g (weigh this accurately, by difference) in ~250 mL of water in a beaker. After the KIO_3 has completely dissolved, quantitatively transfer it to a 500.0 mL volumetric flask. Dilute to the mark and invert the flask 20 - 25 times to mix well (refer to the procedures for preparing a standard solution).

6. Rinse and fill a 50 mL burette with your thiosulphate solution (from step 4).

7. Using a properly prepared pipette, transfer 50.00 mL of the KIO_3 solution to an Erlenmeyer flask.

8. Add 1.3 g of solid KI and 5 mL of 2 mol L^{-1} HCl (neither of these quantities needs to be known accurately).

9. <u>Immediately</u> titrate with the thiosulphate solution until the solution is pale yellow. Add ~2 mL of starch indicator. The solution should turn a deep blue colour.

10. Slowly add thiosulphate until the colour disappears. Careful - the end point is quite sharp and if the starch has been added at the proper time, not much more thiosulphate will be

needed.

11. Repeat the titration with at least two more aliquots of KIO_3 solution or until the relative standard deviation in the thiosulphate concentration is less than 0.3%. Be careful not to run out as you will need at least 150 mL of the KIO_3 solution for part II.

Part II: Analysis of Vitamin C

The solution of the vitamin C unknown should not be prepared until just before beginning the titrations. Aqueous vitamin C is slowly oxidized when exposed to air. However, at the concentrations used in this experiment vitamin C is stable for at least 2 hours. You should complete your titrations within two hours of preparing the vitamin C solutions.

1. Obtain three clean 250 mL Erlenmeyer flasks.

2. To each flask, add ~1.3 g of KI and, using a properly prepared pipette, 50.00 mL of the standard KIO_3 solution. Stopper the flasks.

3. Thoroughly rinse the pipette with deionized water (the pipette will be used below to transfer aliquots of the vitamin C solution).

4. Obtain a vial containing a vitamin C unknown. Be sure to record the code associated with the unknown and the total mass of material in the vial. *Without these data, the results of your analysis will be pointless and you will receive a grade of zero for the laboratory report.*

5. Transfer the entire contents of the vial to a 250 mL beaker. To ensure all the mixture has been transferred, rinse the vial several times with deionized water, transferring the rinsings to the beaker.

6. Add sufficient water to the beaker to the bring the volume to ~125 mL.

7. Stir the solution until all the solid dissolves. Do not remove the stirring rod without rinsing it into the solution.

8. Quantitatively transfer the solution to a 250.0 mL volumetric flask and fill the flask to the mark with deionized water. Invert the flask 20 - 25 times to mix well.

9. Using a properly prepared 25.00 mL pipette, transfer 25.00 mL of the vitamin C solution to one of the Erlenmeyer flasks prepared in step 2.

10. Add 20 mL (graduated cylinder) of 2 mol L^{-1} HCl to the Erlenmeyer flask. The solution should turn a deep reddish brown indicating the presence of excess I_3^-.

11. Immediately titrate the solution with the standardized sodium thiosulphate from part I. Add the starch indicator just before the end point is reached (when the solution is pale yellow). Add thiosulphate until the colour is completely discharged.

12. Repeat the analysis (from step 9) at least two more times.

For CHEM 2130 students: If time permits, you should prepare the glucose working standard solution for "Determination of Glucose", experiment 7.

Results and Calculations

1. Calculate and report the average mass of vitamin C per gram of mixture. Also report a relative standard deviation.

6. MASS TITRATION - VARIATION ON AN OLD THEME

Objectives

1) To determine the concentration of an acetic acid solution.

Introduction

Traditionally, titrations have been done using volumetric glassware and considering concentrations in terms of moles (or mass) of material per unit volume of solution. However, there is no theoretical reason why titrations cannot be carried out on a mass basis. That is, masses are measured instead of volumes and concentrations are defined in terms of moles (or mass) of material per unit *mass* of solution:

$$C_{kg} = \frac{n(solute)}{m(solution)} \tag{6.1}$$

The subscript "kg" is added to emphasize that the units are mol (kg solution)$^{-1}$. The stoichiometric calculations are done in just the same way as with a volumetric titration except that volumes are replaced with masses and molarities (mol L^{-1}) are replaced with mol (kg solution)$^{-1}$.

One great advantage to thinking of solutions in this way is that the concentration of the analyte does not change with temperature. Molarity, on the other hand, is temperature dependent because solution density changes with temperature causing the volume to change. As an aside, if the density of the solution is known (or can be measured), conversion between the two units (mol L^{-1} and mol (kg solution)$^{-1}$) is an easy task. A second advantage to the mass titration is its potential for greater precision because an analytical balance can easily provide 5 - 6 significant figures as

opposed to the 3 - 4 that are typical with a volumetric analysis.

Titrations were originally designed to be done on a volumetric basis because of the relative ease of building and using volumetric glassware versus building and using early balances. However, the proliferation of easy-to-use electronic balances has overcome this advantage and there is now no reason not to perform mass titrations.

In this experiment a mass titration will be used to determine the concentration (mol (kg of solution)$^{-1}$) of an acetic acid solution. In an earlier experiment the concentration of HCl was determined using the primary standard 'tris' (a weak base). Because acetic acid is a weak acid, it cannot be standardized with 'tris' - the pH change at the equivalence is not large enough to generate a "sharp" end point with an appropriate indicator. The acetic acid must be standardized using a strong base such as sodium hydroxide:

$$CH_3COOH(aq) + NaOH(aq) \rightarrow NaCH_3COO(aq) + H_2O(l) \qquad (6.2)$$

Unfortunately, NaOH cannot be used as a primary standard, so it will have to be standardized (also on a mass basis) using a potassium hydrogen phthalate (which <u>is</u> a primary standard) solution:

$$C_8H_5O_4K(aq) + NaOH(aq) \rightarrow C_8H_4O_4NaK(aq) + H_2O(l) \qquad (5.3)$$

Pre-lab Calculations

1. Using the experimental data in the Excel spreadsheet posted on Blackboard, calculate the concentration of acetic acid in the unknown solution. Report a relative standard deviation.

Procedure

Obtain three syringes and needles and label one of the syringes as "KHP", a second as

"NaOH", and the third as "acetic acid". Be very careful handling the needles and syringes, particularly while capping and uncapping the needles. The needles are "blunt", but there is still a risk of a "stick" injury. If you cut yourself with a needle, immediately flush the area with plenty of cool running water and apply a suitable dressing.

In parts II and III repeat the titrations at least three times or until satisfactory agreement is achieved (relative standard deviation less than 0.5%).

Part I: Preparation of ~0.1 mol (kg solution)$^{-1}$ Potassium Hydrogen Phthalate (KHP)

1. Remove your KHP from the oven and store it in a dessicator. Allow it to cool thoroughly (15 - 20 minutes).

2. Use the weight-by-difference technique to transfer an accurately known mass (~3 g) of KHP to a 250 mL beaker containing a stir bar.

3. Record the mass of the beaker + KHP (use an open-pan balance).

4. Add ~150 mL of deionized water to the beaker and stir (not too fast to avoid spattering) until all the KHP has dissolved (this may take several minutes).

5. Once the KHP has completely dissolved, wash down the sides of the beaker with deionized water and continue to stir the solutions for a few minutes. Record the mass of the beaker and solution (use an open-pan balance) and calculate the mass of water added.

6. Cover the beaker with a watch glass.

Part II. Preparation and Standardization of ~0.1 mol (kg solution)$^{-1}$ NaOH

1. Obtain ~5 mL of 12.5 mol (kg solution)$^{-1}$ NaOH in a 10 mL graduated cylinder. Pour the 5 mL into a 500 mL plastic bottle. Thoroughly rinse the cylinder with deionized water (because the NaOH will react with the glass and damage it).

   Caution: 12.5 mol (kg solution)$^{-1}$ NaOH is very caustic. Handle it with care. In the event of skin exposure, rinse thoroughly with water. For eye exposure, flush for 30 minutes at the eyewash station and seek medical attention.

2. Add ~500 mL of boiled deionized water to the plastic bottle. Cap tightly and mix well.

3. Fill the syringe marked "KHP" with ~15 mL of the KHP solution. Cap the needle (be *very* careful) and weigh (in a beaker or flask) the syringe and needle on the analytical balance. Record the mass (4 decimal places).

4. Discharge the contents of the syringe into a 125 mL Erlenmeyer flask containing a stir bar. Avoid any drops adhering to the tip of the needle. Cap the needle and record the mass of the now empty syringe (use the same analytical balance as in step 3).

5. Add 2 - 3 drops of phenolphthalein indicator to the solution in the Erlenmeyer flask. Place the Erlenmeyer on a magnetic stirrer and start the stirrer (not too fast, to avoid splashing).

6. Decant ~100 mL of the ~0.1 mol (kg solution)$^{-1}$ NaOH solution into a clean, rinsed (with the NaOH solution) 250 mL beaker. Fill the syringe labelled "NaOH" with ~20 mL of the NaOH solution. Cap the needle, place the syringe in a small beaker and record the mass of the syringe + beaker (analytical balance).

7. Slowly add NaOH from the syringe to the potassium hydrogen phthalate solution in the 125 mL Erlenmeyer flask until the solution turns a persistent, faint pink. Initially, the solution will turn pink, but the colour will quickly disappear with stirring. As the end point is approached, the colour will persist longer and longer. As you get closer to the end point, wash down the sides of the flask with deionized water to ensure all the NaOH reacts with the KHP solution. Once the colour persists for ~30 s, the end point has been reached. Stop adding NaOH.

8. Cap the NaOH syringe and re-weigh it (in the same beaker used in step 6) on the analytical balance. Calculate the mass of NaOH solution added to the Erlenmeyer flask.

9. For each titration calculate the concentration of NaOH in mol (kg solution)$^{-1}$. Repeat the titrations (steps 3 - 8) until sufficient agreement is reached (three titrations for which the relative standard deviation is less than 0.5%). Average these values and use this average concentration for your calculations.

Part III: Determination of the Concentration of Acetic Acid

1. Obtain ~150 mL of ~ 0.1 mol (kg solution)$^{-1}$ acetic acid in a clean, dry beaker from the stock bottle at the front of the lab. The beaker should be dry to avoid diluting the acetic acid.

2. Repeat the procedure in Part II using the unknown acetic acid instead of KHP. Use the third syringe to measure the aliquots of acetic acid.

At the end of the experiment dispose of the needles in the receptacle provided. Clean and thoroughly rinse all glassware.

If time permits, you should prepare the glucose working standard solution for "Determination

of Glucose", experiment 7.

Results and Calculations

1. Calculate the C_{kg} of the potassium hydrogen phthalate solution.

2. Calculate the average C_{kg} of your NaOH solution. Report a relative standard deviation.

3. Calculate the average C_{kg} of the acetic acid unknown. Report a relative standard deviation.

7. DETERMINATION OF GLUCOSE CONCENTRATION BY AN INDIRECT SPECTROPHOTOMETRIC METHOD

Objective

1) To illustrate the application of the Beer - Lambert Law (or Beer's Law) to chemical analysis; 2) To determine the concentration of glucose in an aqueous sample.

Introduction

Glucose ($C_6H_{12}O_6$) is a "simple" sugar (or monosaccharide) formed by plants from carbon dioxide and water using energy from sunlight (photosynthesis). Biological cells metabolize glucose and use the energy released to drive the biochemical processes of life. In effect, solar energy is stored as chemical energy in the molecules of glucose. Both plants and animals store glucose (in polymeric form) for future needs: in plants it is stored as starches and in animals it is stored as glycogen.

In human beings glucose is transported to biological cells through the bloodstream and the concentration of glucose in the blood must be strictly regulated. The body regulates glucose concentration in blood by producing the hormone insulin. Diabetes mellitus is a collection of metabolic diseases in which the concentration of glucose in the blood is not properly regulated. Diabetes can result if the body does not produce sufficient insulin (type 1 diabetes) or if the insulin produced is ineffective at regulating the glucose levels (type 2). The long term health consequences of untreated diabetes can be severe: cardiovascular disease, kidney failure, blindness, etc.

Reliable determination of blood glucose levels is of great clinical importance for both the

diagnosis and treatment of diabetes. Glucose levels in blood can be measured using either electrochemical methods (the basis for many glucometers used for "at home" glucose monitoring) or spectrophotometric methods based on the Beer - Lambert Law (the basis for glucose measurements done in medical testing laboratories).

In simple spectrophotometric analyses the concentration of analyte in a sample can be determined from the light absorbed (at a given wavelength) by the analyte using a linear relationship (the Beer-Lambert Law):

$$A = \varepsilon b C \tag{7.1}$$

where: A = absorbance (the amount of light absorbed)

ε = molar absorptivity (L mol^{-1} cm^{-1})

b = path length (cm)

C = concentration of the absorbing species (mol L^{-1})

The molar absorptivity is a proportionality constant and is characteristic of the absorbing species. It depends on the wavelength of the incident light and indicates how well a given wavelength is absorbed by the analyte. Normally, to minimize error, a wavelength is chosen where ε is a maximum. The Beer-Lambert Law predicts a linear relationship between absorbance and concentration, but this is only true when the concentration of the absorbing species is comparatively low; at higher concentrations the relationship becomes non-linear.

In theory, if the path length and molar absorptivity were known, it would be possible to calculate the concentration from (see experiment 3):

$$C = \frac{A}{\varepsilon b} \tag{7.2}$$

In practice, however, this is generally not done. The unknown concentration is determined by interpolation using a "standard curve". The standard "curve" is most often a line and it is

constructed by plotting the absorbance (y-axis) versus concentration (x-axis) data for a number of solutions with very accurately known concentrations of the species being analyzed. A line is fit to the data using linear regression and the equation for the line is used to calculate the unknown concentrations from the measured absorbances of the samples. For greatest accuracy the unknown concentrations should lie somewhere between the lowest and highest concentration of the standards (optimally, in the middle of the range). The more standards used to construct the plot the better because the fitted function will give statistically better estimates of the slope and intercept of the line.

Unfortunately, glucose does not absorb light (it is colourless both in pure form and in aqueous solution), so an indirect approach to measuring its concentration spectrophotometrically must be used. This is frequently done by reacting the glucose with some reagent(s) to produce a chemical species that *does* absorb light (the "chromophore"). In order for the analysis to work the concentration of the chromophore must be directly proportional to the concentration of glucose. The "Trinder Glucose Activity" test is based on this methodology and is widely used for measuring glucose levels in blood. In the Trinder test glucose in a blood sample is oxidised (by glucose oxidase) to hydrogen peroxide and gluconic acid. In the presence of peroxidase the hydrogen peroxide reacts with phenol and 4-aminoantipyrine to form quinoneimine which is the chromophore. The concentration and absorbance (the measured quantity) of the quinoneimine are directly proportional to the concentration of glucose in the blood sample.

In this experiment the chromophore is $Fe(CN)_6^{3-}$ and the chemistry is simpler than the Trinder test. Glucose is a "reducing sugar" meaning it is relatively easily oxidised (good for physiology!) and it will reduce the oxidation state of other species. Glucose reacts with $Fe(CN)_6^{3-}$ according to:

$$\text{Glucose} + a\ Fe(CN)_6^{3-} \rightarrow a\ Fe(CN)_6^{4-} + \text{other products} \qquad (7.3)$$

where 'a' is the constant (but unknown) stoichiometric ratio between glucose and $Fe(CN)_6^{3-}$. In this system, the only species that absorbs light is $Fe(CN)_6^{3-}$. Because reaction (7.3) *reduces* the concentration of $Fe(CN)_6^{3-}$, the higher the concentration of glucose, the *lower* the absorbance will be. The absorbance is given by:

$$A = \varepsilon b\left[Fe(CN)_6^{3-}\right] \tag{7.4}$$

In this experiment a series of solutions will be prepared containing a *fixed* initial concentration of $Fe(CN)_6^{3-}$ and variable initial concentration of glucose. Under these conditions it can be shown that:

$$A = d + k\,F_{glucose} \tag{7.5}$$

where 'd' and 'k' are constants and $F_{glucose}$ is the "formal" concentration of glucose (i.e., the concentration of glucose neglecting any reaction with $Fe(CN)_6^{3-}$). In other words, the absorbance is linearly dependent on the formal concentration of glucose. To analyse the glucose in the test solution the same amount of $Fe(CN)_6^{3-}$ is combined with a volume of a solution of unknown glucose concentration and the absorbance is measured. Using equation (7.5), the concentration of the glucose in the test solution can be calculated from the observed absorbance. This simpler method (compared to the Trinder test) is used because the samples in this experiment will be simple aqueous solutions as opposed to blood samples.

Prelab Questions

1. Starting with equation (7.4) derive equation (7.5). *Hint: write $[Fe(CN)_6^{3-}]$ in terms of the formal concentration of glucose and the initial concentration of $Fe(CN)_6^{3-}$ and recognize that 'd' and 'k' are arbitrary constants.*

2. Using the experimental data in the Excel spreadsheet posted on Blackboard, calculate the concentration of glucose in the *original* solution. Report the relative standard deviation.

Procedure

Prior to performing this experiment you should review: weight by difference, the volumetric flask, the transfer pipette, the Mohr pipette, preparing a standard solution and the Genesys20 spectrophotometer. Proper mixing of solutions is vitally important.

A solution containing ~6.0 mmol L^{-1} $K_3Fe(CN)_6$ and 0.5 mol L^{-1} Na_2CO_3 will be provided. Note the concentration of the $K_3Fe(CN)_6$.

Part I: Preparation of Standard Glucose Solution

1. Using the weight-by-difference technique, transfer ~0.62 g (accurately known) of glucose to a 250 mL beaker.

2. Dissolve the glucose in ~125 mL of deionized water. Quantitatively transfer this solution to a 250.0 mL volumetric flask and dilute the resulting solution to mark with deionized water. Invert the flask 20 - 25 times to mix well. This is the stock standard solution of glucose and the concentration should be ~2 500 mg L^{-1}.

3. Using a properly prepared 5.00 mL transfer pipette, transfer 5.00 mL of the stock standard solution of glucose to a second 250.0 mL volumetric flask. Dilute to the mark with deionized water and invert the flask 20 - 25 times to mix well. This is the working standard solution of glucose and the concentration should be ~50 mg L^{-1}. This solution will be used for the experiment.

4. If the working standard solution must be stored to use in a later laboratory period, thoroughly clean and rinse a polyethylene bottle (of suitable size) and transfer the working standard to it.

Part II: Determination of Glucose Concentration

Reaction (7.3) needs to be heated to proceed at a reasonable rate. While preparing the solutions below, heat roughly 250 mL of tap water to ~60^0C on a hot plate.

1. Obtain 9 clean, but not necessarily dry, 20 x 150 mm test tubes. Label them 1 - 9.

2. Collect ~70 mL of the $K_3Fe(CN)_6$ solution in a clean, dry beaker. The beaker must be dry to avoid diluting the solution.

3. Using a properly prepared transfer pipette transfer 5.00 mL of the $K_3Fe(CN)_6$ solution to each of test tubes 1 - 9. It is *very* important that exactly the same amount of the $K_3Fe(CN)_6$ solution is transferred to each test tube. This ensures the same initial concentration of $Fe(CN)_6^{3-}$ in each of the analysed solutions; a necessary condition of the method.

4. Using a properly prepared Mohr pipette transfer the following (nominal) volumes of the glucose working standard solution to the first six test tubes (note the <u>exact</u> volume transferred in each case):

Test Tube	Volume of glucose solution / mL
1	0
2	2
3	3
4	6
5	7
6	9

Note: all six transfers can be completed with only three fills of the Mohr pipette!

5. Collect ~30 mL of your assigned glucose solution in a clean, dry beaker. Again, the beaker must be dry to avoid diluting the solution.

6. Using a properly prepared 5.00 mL transfer pipette, transfer 5.00 mL of the unknown glucose solution to each of test tubes 7 - 9.

7. Cover each test tube well with Parafilm and invert a few times to mix well.

8. Place test tubes 1 - 9 in a 1 L beaker. Add a tenth test tube containing ~10 mL of water. Carefully add warm water to the 1 L beaker until the solutions in the test tubes are completely submerged.

9. Add a couple of boiling stones and heat the 1 L beaker on a hot plate until the water boils. Reduce the heat (but maintain boiling) and let the reaction run for 15 minutes.

10. After 15 minutes carefully remove the beaker from the hot plate and transfer it to the sink. Be careful - the beaker will be hot. Only handle the beaker at a level above the water in the beaker and preferably only at the very top. If the beaker is too hot to handle, use beaker tongs.

11. Set up a gentle to moderate flow of water through the rubber hose in the sink. Carefully insert the hose into the 1 L beaker (below the level of the water) and allow the running water to "flush" the 1 L beaker. Do not allow any water to enter the test tubes. Periodically, place a thermometer in the 10^{th} test tube (the one containing only water) and measure the temperature of the water.

12. Once the temperature in the 10^{th} test tube is 20 - 25^0C, turn off the flow of water and remove test tubes 1 - 9 from the beaker. Quantitatively transfer the contents of each test tube to separate 50.0 mL volumetric flasks. Dilute each solution to the mark and invert 20 - 25 times to mix well.

13. Measure the absorbance at 420 nm of each of the solutions. Use deionized water as the blank. Measure solutions 6 to 1 (in that order) and then measure the three replicate unknowns. Between each sample, rinse the cuvette once with deionized water and then three times with the next solution to be measured.

Results and Calculations

1. Determine the concentration of glucose (mg L^{-1}) in the *original* glucose solution. Report a relative standard deviation for the original concentration.

8. DETERMINATION OF pK_a OF AN ACID - BASE INDICATOR

Objectives

1) To calculate the quantities of conjugate acid and of sodium hydroxide required to prepare a buffer of a given pH; 2) To determine the pK_a of an acid - base indicator.

Introduction

An acid - base indicator is a weak acid that has one colour when it is protonated (at lower pH) and different colour when it is deprotonated (at higher pH). The different colours imply the absorbance (at a given wavelength) of the indicator depends on the pH. This is because the ratio of $[In^-]$:[HIn] depends on the pH. If the absorbance is measured as a function of pH, the ratio $[In^-]$:[HIn] can be found as a function of pH. From this the pK_a can be calculated.

The dissociation reaction for a generic indicator can be written as:

$$HIn(aq) \rightleftarrows H^+(aq) + In^-(aq) \tag{8.1}$$

For this reaction an equilibrium constant can be written[3]:

$$K_a = \frac{\left[H^+\right]\left[In^-\right]}{\left[HIn\right]} \tag{8.2}$$

From this it is fairly simple to show:

[3] *Strictly speaking the thermodynamic equilibrium constant, K_a, should be defined in terms of activities. In this experiment we are neglecting activity coefficient (assuming they are all equal to 1). As such the pK_a found in this experiment is an approximation of the true, thermodynamic pK_a.*

$$\log_{10}\frac{[In^-]}{[HIn]} = pH - pK_a \tag{8.3}$$

A plot of $\log_{10}\frac{[In^-]}{[HIn]}$ versus pH should be linear with slope = 1 and intercept = $-pK_a$. Therefore, if the ratio of the concentrations can be measured as function of pH, the pK_a of the indicator can be found.

In this experiment $\frac{[In^-]}{[HIn]}$ will be found from absorbance data. Generally, In^- and HIn have different wavelengths of maximum absorbance, λ_{max} (because they have different colours). In the case of bromothymol blue $\lambda_{max}(In^-) = 615$ nm and $\lambda_{max}(HIn) = 432$ nm. Let us assume that HIn and In^- are the only two species in the system that absorb light. If this is true, then at any given wavelength, λ, the observed absorbance, A_λ, can be written as the sum of the absorbances of HIn and In^-:

$$A_\lambda = A_{In^-} + A_{HIn} \tag{8.4}$$

Making use of Beer's Law:

$$A_\lambda = \varepsilon_{In^-,\lambda} b[In^-] + \varepsilon_{HIn,\lambda} b[HIn] \tag{8.5}$$

where $\varepsilon_{In^-,\lambda}$ = the molar absorptivity if In^- at λ

$\varepsilon_{HIn,\lambda}$ = the molar absorptivity if HIn at λ

(Note: $\varepsilon_{In^-,\lambda}$ and $\varepsilon_{HIn,\lambda}$ depend on the wavelength, λ)

b = the path length of the cuvette

If HIn and In^- are in equilibrium, then mass balance requires:

$$F = [HIn] + [In^-] \tag{8.6}$$

where 'F' is the "formal" concentration of the indicator. If a path length of 1 cm is used, it can be shown that (see question 3):

$$\frac{[In^-]}{[HIn]} = \frac{\varepsilon_{HIn,\lambda} F - A_\lambda}{A_\lambda - \varepsilon_{In^-,\lambda} F} \tag{8.7}$$

To apply this equation the two molar absorptivities must be known as well as the formal concentration of the indicator. The formal concentration of the indicator can be calculated from amount of indicator used to prepare the solution. The molar absorptivity of HIn at λ can be found by measuring the absorbance of a solution of accurately known concentration of HIn. However, because the HIn is in equilibrium with In^-, preparing such a solution is difficult. This problem is solved by driving the equilibrium towards HIn by increasing the $[H^+]$ (lowering the pH). So, a known amount of indicator is dissolved and then the pH is lowered to an "extreme" value and the absorbance is measured. At the "extreme" pH all of the HIn is assumed to exist in the protonated form, so [HIn] is known. The absorbance at the wavelength of interest is measured and the molar absorptivity of HIn is calculated. Likewise, the molar absorptivity of In^- can be found by measuring the absorbance of a solution with a very high pH (because the high pH ensures the HIn is completely deprotonated).

However, with good experimental design it is possible to measure $\frac{[In^-]}{[HIn]}$ *without* explicitly finding the two molar absorptivities and without knowing the exact formal concentration of the indicator. In this experiment a series of solutions will be prepared with a <u>constant</u> formal concentration of the indicator (although this concentration will not be accurately known). The series of solutions will have one with very low pH (the "acid extremum"), one with a very high pH (the

"base extremum") and the remainder with moderate pH's. Again, consider HIn. At a given wavelength, λ, the molar absorptivity of HIn is constant. If the formal concentration of the indicator is constant in all the solutions, the term:

$$\varepsilon_{HIn,\lambda} \times F \tag{8.8}$$

is constant for all the solutions. Recall that in equation (8.7) a path length of 1.00 cm was assumed. Now we can write (from the Beer - Lambert Law):

$$A_{HIn,\lambda} = \varepsilon_{HIn,\lambda} \times 1.00\,cm \times F = \varepsilon_{HIn,\lambda} \times F \tag{8.9}$$

where $A_{HIn,\lambda}$ is the absorbance of HIn (at λ) in the acid extremum solution. $A_{HIn,\lambda}$ is constant as long as F is constant. In a similar way:

$$A_{In^-,\lambda} = \varepsilon_{In^-,\lambda} \times 1.00\,cm \times F = \varepsilon_{In^-,\lambda} \times F \tag{8.10}$$

where $A_{In^-,\lambda}$ is the absorbance of In (at λ) in the base extremum solution. $A_{In^-,\lambda}$ is also constant as long as F is constant. Therefore, we can re-write equation (8.7) as:

$$\frac{[In^-]}{[HIn]} = \frac{A_{HIn,\lambda} - A_\lambda}{A_\lambda - A_{In^-,\lambda}} \tag{8.11}$$

where $A_{HIn,\lambda}$ and $A_{In^-,\lambda}$ are the absorbances in the acid extremum and base extremum solutions, respectively. This allows us to find the ratio $\frac{[In^-]}{[HIn]}$ in each solution of different pH from the absorbance (at λ), A_λ, of that solution.

To measure the pK_a of the indicator, a series of solutions containing a fixed amount of indicator at different pH's is prepared. The pH of each solution in the series must remain constant which is accomplished by "buffering" the pH of the solution. Using buffers also allows solutions with small differences in pH to be prepared easily.

pH buffers are prepared by mixing a weak acid with its conjugate base (for example, acetic

acid and the acetate ion). If a strong base (OH^-) is added to the buffer, it reacts with the weak acid, reducing the concentration of conjugate acid and increasing the concentration of weak base. The pH will increase slightly, but not as much as if the NaOH were added to deionized water. If a strong acid is added to a buffer, the reverse process occurs: conjugate base is consumed and conjugate acid is produced, but the pH changes only slightly. A buffer's resistance to pH change depends on the amount of each of the conjugate base and conjugate acid present and on their ratio; for a fixed total amount of acid plus conjugate base the maximum buffering is achieved when the ratio of conjugate base to conjugate acid is one.

The pH of a buffer is governed by the ratio of weak base concentration to weak acid concentration (the Henderson - Hasselbalch equation):

$$pH = pK_a + \log_{10} \frac{[CH_3COO^-]}{[CH_3COOH]} \tag{8.12}$$

Because the conjugate acid and base are contained in the same volume, the Henderson - Hasselbalch equation can be re-written as:

$$pH = pK_a + \log_{10} \frac{n_{CH_3COO^-}}{n_{CH_3COOH}} \tag{8.13}$$

Instructions for preparing a buffer will always state the desired pH and will specify either the concentration of one of the components (the conjugate acid or the conjugate base) or the sum of the concentrations of the conjugate acid and conjugate base. Because a buffer has its maximum effectiveness when the ratio of the conjugate base to conjugate acid is one (and, therefore, $pH = pK_a$), it is best to choose a weak acid (or base) whose pK_a is as close as possible to the intended pH of the buffer. Of course, other factors must be considered: availability, ease of handling, stability of reagents, etc.

Buffers can be prepared in one of two ways. The easiest method is to mix appropriate

amounts of the acid and the conjugate base together in a vessel. Diluting a buffer has only a small effect on the pH (for reasonable amounts of water), so water is added to the mixture to prepare the desired volume. The second method is to begin with the weak acid (or weak base) and add strong base (strong acid) such as sodium hydroxide (hydrochloric acid). The hydroxide reacts with the acid and partially converts it to the conjugate base. This creates a system which is a mixture of the conjugate acid and its conjugate base:

$$CH_3COOH(aq.) + NaOH(aq.) \rightarrow NaCH_3COO(aq.) + H_2O(l) \qquad (8.14)$$

and creates the buffer system. When starting with the base, the HCl reacts with the weak base to produce the conjugate acid.

The second method is not any more difficult, but does require more calculations and more thought! Perhaps an example is in order.

Starting with 1.0 mol L^{-1} CH_3COOH and 1.0 mol L^{-1} NaOH, how would you prepare 500 mL buffer that has a pH of 5.00 and total concentration of acetic acid and sodium acetate ($[CH_3COOH] + [CH_3COO^-]$) of 0.50 mol L^{-1}?

The key to buffer problems is determining the ratio of the numbers of moles in the Henderson-Hasselbalch equation or finding the individual numbers of moles of the conjugate acid and base. For the latter you need to find the two unknowns (n_{HA} and n_{A^-}), so you need two equations (representing two conditions). In this example the two conditions are i) the total number of moles ($n_{CH_3COOH} + n_{CH_3COO^-}$) which is expressed through the total (formal) concentration and ii) the ratio $n_{CH_3COO^-} / n_{CH_3COOH}$ expressed through the pH and the Henderson - Hasselbalch equation.

In this problem we are told that $[CH_3COOH] + [CH_3COO^-] = 0.50$ mol L^{-1}. Multiplying by the volume of the buffer (0.500 L) gives:

$$n_{CH_3COOH} + n_{CH_3COO^-} = 0.250\,mol \tag{8.15}$$

This is the first equation. The second equation is the Henderson - Hasselbalch equation:

$$pH = pK_a + \log_{10}\frac{n_{CH_3COO^-}}{n_{CH_3COOH}} \tag{8.16}$$

Equation (8.15) can be re-arranged and substituted into equation (8.16) to give:

$$5.0 = 4.75 + \log_{10}\frac{n_{CH_3COO^-}}{0.250 - n_{CH_3COO^-}} \tag{8.17}$$

Solving this latter equation for $n_{CH_3COO^-}$ gives the number of moles of acetate required *in the buffer*; $n_{CH_3COO^-}$ = 0.160 mol. The acetate is produced <u>only</u> by the reaction of the acetic acid with NaOH (which is the limiting reagent) and the stoichiometry is 1:1. To make 0.160 mol of CH_3COO^-, 0.160 mol of NaOH must be added. This requires 160 mL of 1.0 mol L^{-1} NaOH. How much acetic acid is needed, initially? It must be the amount that is in the buffer n_{CH_3COOH}, plus the amount of acetic acid that reacted with the NaOH:

$$n_{CH_3COOH,i} = n_{CH_3COOH} + n_{CH_3COOH,reacted} \tag{8.18}$$

The stoichiometry of reaction (8.14) tells us that the $n_{CH_3COOH,reacted}$ is the same as $n_{CH_3COO^-}$, so:

$$n_{CH_3COOH,i} = n_{CH_3COOH} + n_{CH_3COO^-} \tag{8.17}$$

Using equation (8.15) we can write:

$$n_{CH_3COOH,i} = 0.250\,mol \tag{8.18}$$

Hence, the buffer is prepared by mixing 250 mL of 1.0 mol L^{-1} acetic acid with 160 mL of 1.0 mol L^{-1} NaOH and adding 90 mL of deionized water to make the volume up to 500 mL.

In this experiment the indicator is bromothymol blue. The solutions of bromothymol blue will be buffered at pH's between ~6.3 and ~8.2 using a $H_2PO_4^-$ / HPO_4^{2-} buffer ($pK_{a,2}$ = 7.2). The buffers are prepared by mixing KH_2PO_4 and NaOH.

Procedure

Prior to beginning this experiment you should review the instructions on the transfer pipette, volumetric flask, preparing a standard solution, the Genesys20 spectrophotometer and using the Seven Easy pH meter.

Pre-lab Calculations

1. Calculate the mass of KH_2PO_4 required to prepare 100 mL of a solution in which the formal concentration of $H_2PO_4^-$ ($F(H_2PO_4^-)$) is 0.20 mol L^{-1}.

2. Calculate the volume of 0.20 mol L^{-1} KH_2PO_4 and the volume of 0.10 mol L^{-1} NaOH required to prepare 50.0 mL of buffer with a formal (or total) concentration of $H_2PO_4^-$ of 0.020 mol L^{-1} and a pH = 6.3. Repeat this calculation for buffers (of the same volume and formal concentration) with pH = 6.5, 6.9, 7.5, 7.9, and 8.2. You need only show one sample calculation. You should be able to set up a spreadsheet to do these calculations.

3. When preparing the solutions in the experiment, the protocol requires the bromothymol blue solutions be transferred with a pipette. However, the volumes of NaOH and KH_2PO_4 solutions can be transferred by a graduated cylinder. Why is it not necessary to know these volumes accurately?

Part I: Preparation of Solutions

1. Transfer the mass of KH_2PO_4 calculated in the pre-lab calculations to a clean 250 mL beaker. Using a graduated cylinder, add 100 mL of water and stir to completely dissolve the KH_2PO_4.

2. Collect 8 clean 50.0 mL volumetric flasks and label them 1 through 8.

3. In a clean, dry 250 mL beaker, collect ~100 mL of 0.20 g L^{-1} bromothymol blue solution.

4. Using a properly prepared 10.00 mL transfer pipette, transfer 10.00 mL of the bromothymol blue solution to each of the 8 volumetric flasks.

5. To flask 1 add 10 mL of 0.1 mol L^{-1} HCl. Dilute the solution to the mark with deionized water. Invert the flask 20 - 25 times to mix well. This is the acid extremum.

6. To flask 2 add the volume of KH_2PO_4 solution and the volume of 0.10 mol L^{-1} NaOH solution calculated in step 2 of the pre-lab calculations (the volume required to prepare a buffer at 6.3). A graduated cylinder can be used to measure these volumes. Dilute to the mark with deionized water. Invert the flask 20 - 25 times to mix well.

7. Repeat step 6 with flasks 3 - 7 for the remaining buffer pH values.

8. Add 10 mL of 0.1 mol L^{-1} NaOH to flask 8. Dilute to the mark and invert 20 - 25 times to mix well. This is the base extremum.

Part II: Measurements

One half of the Genesys 20 spectrophotometers in the laboratory will be set to 432 nm and

the other half will be set to 615 nm. For all spectrophotometric measurements in this experiment use deionized water as the blank. You should use the same spectrophotometer for all measurements at a given wavelength.

1. Using a properly rinsed 1 cm cuvette, measure the absorbance of solution 2 at $\lambda = 432$ nm (in the first spectrophotometer) and then at $\lambda = 615$ nm (in the second spectrophotometer).

2. After the absorbance has been measured, decant a portion of solution 2 from the 50.0 mL volumetric flask into a small, clean, properly rinsed beaker and measure the pH of the solution.

3. Repeat steps 1 and 2 with solutions 3 - 7. Between each solution rinse the cuvette once with deionized water and then three times with the next solution to be used.

4. Repeat step 1 with solutions 1 (acid extremum) and 8 (base extremum). There is no need to measure the pH of solutions 1 and 8. Again, rinse the cuvette once with deionized water and three times with the solution to be measured.

5. At the end of the experiment you should have measurements (pH, Absorbance) for six buffered solutions and one absorbance measurement for each of the two extreme pH solutions. You should have measured the absorbance at each of two wavelengths.

Results, Calculations and Questions

The report for this experiment is extensive and the questions are challenging. Plan accordingly!

1. Tabulate your data for the experiment. Show a sample calculation of $\frac{[In^-]}{[HIn]}$.

2. Using the data for λ = 432 nm, create a plot of $\log_{10}\frac{[In^-]}{[HIn]}$ vs. pH and from it determine the pK_a of bromothymol blue. What is the 95% confidence interval for the pK_a? See the Excel guide for more information on calculating the 95% confidence interval. Repeat this calculation for the data for λ = 615 nm. In both case the degrees of freedom is n – 2 where n is the number of points on the line.

3. Beginning with equations (8.5) and (8.6), derive equation (8.7). Assume a path length of 1 cm.

4. The pH of the acid extremum is ~1.7. At this pH show that it is reasonable to assume $[In^-]$ is negligible.

5. Imagine you repeated this experiment with a new pH meter that had a constant error of +p units (that is, it reported a pH of 6.3 + p when the pH was actually 6.3). Assuming the absorbance measurements were the same, would the second pK_a be higher or lower than the first? In terms of p, m (the slope of your plot) and the true pK_a, what would the new (erroneous) pK_a be?

6. The ability of a buffer to resist pH change when strong acid or strong base is added is measured by its "buffer capacity". Buffer capacity, β, can be defined by:

$$\beta = \frac{dC_b}{dpH} = \frac{-dC_a}{dpH} \qquad (8.19)$$

where dC_b and dC_a are the changes in the concentration of conjugate acid or conjugate base as strong monoprotic acid or base are added, respectively. The units of buffer capacity are

"mol L^{-1}". As a consequence of the definitions, buffer capacity is always a positive quantity.

For a monoprotic acid the buffer capacity (as a function of pH) is given by:

$$\beta = \left(H + \frac{K_W}{H} + \frac{K_a F H}{(K_a + H)^2} \right) \times \ln 10 \qquad (8.20)$$

where $H = 10^{-pH}$ and F is the total concentration of acid and conjugate base ($F = [HA] + [A^-]$). The buffer capacity depends on both the pH of the buffer and the concentrations of the components of the buffer.

Calculate the theoretical buffer capacity for an acetic acid / sodium acetate buffer (pK_a = 4.75, F = 0.05 mol L^{-1}, $K_w = 1.0 \times 10^{-14}$) as a function of pH (for values of pH = 3 to pH = 10, with an increment in pH of 0.1 unit) using equation (8.20). Plot theoretical β vs. pH.

7. Neglecting the first two terms in the brackets on the right hand side of equation (8.20), show that when $pH = pK_a$, the buffer capacity is given by:

$$\beta = 0.576F \qquad (8.21)$$

where F is the total concentration of conjugate acid and conjugate base. At this pH the buffer capacity is maximized.

8. *(Bonus question for Chemistry 2130 students; required for Chemistry 2030 students).* Starting with equation (8.20) show that the maximum buffer capacity occurs when $pH = pK_a$. If you use Maple (or any other computer programme), include the relevant code. The first two terms in the brackets on the right hand side of equation (8.20) can be neglected.

9. GRAN TITRATION TO DETERMINE CONCENTRATION OF ACETIC ACID

Objective

1) To determine the unknown concentration of an acetic acid solution.

Introduction

In an acid - base titration the objective is to find the equivalence point (i.e., the volume of titrant required to cause the acid and the base to be present in their stoichiometric ratio, often 1:1). The equivalence point is estimated in two common ways. The first is to use an indicator which (most often) changes colour at the equivalence point. Strictly, this method gives the end point, but if the indicator is well-chosen, the difference between the end point and the equivalence point is negligibly small. Using indicators is fast, but can be subject to bias or error on the part of the analyst because of their perception of colour changes. The second common method is to measure the pH as a function of the volume of added titrant. The simplest approach is to plot dpH / dV (usually approximated as ΔpH / ΔV) and to determine the volume at which ΔpH / ΔV is a maximum (c.f. pH titration lab in first-year chemistry). This volume is the volume required to reach the equivalence point. A more sophisticated approach (for titrating a weak acid with a strong base) is to construct a Gran Plot and determine the equivalence point from it. A Gran Plot is a graph of $V_B \times 10^{-pH}$ vs. V_B (where V_B is the volume of strong base added). The equivalence point is found from the x-intercept of this plot.

For the dissociation of a weak acid, HA:

$$HA(aq) \rightleftharpoons H^+(aq) + A^-(aq) \qquad (9.1)$$

a thermodynamic equilibrium constant can be written as:

$$K_a = \frac{[H^+]\gamma_{H^+}[A^-]\gamma_{A^-}}{[HA]\gamma_{HA}} \tag{9.2}$$

where: [i] = the equilibrium concentration of the i^{th} species

γ_i = the activity coefficient of the i^{th} species.

Now, consider the circumstance of a titration in which a strong base (e.g. NaOH) is added to the weak acid. The acid and the base react according to:

$$HA(aq) + OH^-(aq) \rightarrow A^-(aq) + H_2O(l) \tag{9.3}$$

If the sample is initially just HA, the only source of A^- comes from the reaction of the HA with the added NaOH (assuming the amount of A^- from reaction (9.1) is negligible which is reasonable for small K_a). At any point in the titration the formal concentration of A^-, F_{A^-}, can be calculated from the stoichiometry of the reaction:

$$F_{A^-} = \frac{n_{A^-,produced}}{V_{tot}} = \frac{n_{B,added}}{V_{tot}} = \frac{V_B C_B}{V_B + V_{HA}} \tag{9.4}$$

where: C_B = the formal concentration of the strong base (the titrant)

V_B = the volume of added base (the titrant volume)

V_{HA} = the initial volume of acid

(Note: this equation only applies when the stoichiometry is 1:1 between acid and base). The formal concentration of HA, F_{HA}, after an addition of NaOH will be given by:

$$F_{HA} = \frac{n_{HA,remain}}{V_{tot}} = \frac{C_{HA}V_{HA} - C_B V_B}{V_{HA} + V_B} \tag{9.5}$$

where C_{HA} = the <u>initial</u>, formal concentration of the acid

If the acid is sufficiently weak (i.e., the equilibrium in equation (9.1) lies far to the left; K_a

is very small), $[HA] \approx F_{HA}$ and $[A^-] \approx F_{A^-}$ and the expressions for F_{HA} and F_{A^-} can be substituted into equation (9.2) to yield:

$$K_a = \frac{[H^+]\gamma_{H^+} V_B C_B \gamma_{A^-}}{(V_{HA}C_{HA} - V_B C_B)\gamma_{HA}} \tag{9.6}$$

This is easily re-arranged to give:

$$V_B[H^+]\gamma_{H^+} = \frac{K_a \gamma_{HA}}{\gamma_{A^-}}\left(\frac{V_{HA}C_{HA}}{C_B} - V_B\right) \tag{9.7}$$

The number of moles of base required to reach the equivalence point is given by:

$$n_{B,eq} = C_B V_e \tag{9.8}$$

V_e is the volume of base required to reach the equivalence point, the objective of the experiment. Because the stoichiometry is 1:1:

$$n_{B,\,eq} = n_{HA,i} = C_{HA}V_{HA} \tag{9.9}$$

and

$$V_e = \frac{C_{HA}V_{HA}}{C_B} \tag{9.10}$$

equation (9.7) can be written as:

$$V_B 10^{-pH} = \frac{K_a \gamma_{HA}}{\gamma_{A^-}}\left(V_e - V_B\right) \tag{9.11}$$

For dilute solutions we may assume $\gamma_{HA} / \gamma_{A^-}$ is nearly constant (as is K_a). V_e is also constant, so equation (9.11) can be written as:

$$V_B 10^{-pH} = \frac{K_a \gamma_{HA}}{\gamma_{A^-}}V_e - \frac{K_a \gamma_{HA}}{\gamma_{A^-}}V_B \tag{9.12}$$

$$V_B \times 10^{-pH} = b + mV_B \tag{9.13}$$

and it is the form of a line with $y = V_B \times 10^{-pH}$ and $x = V_B$. Therefore, a plot of $V_B \times 10^{-pH}$ versus V_B (the "Gran Plot") should be linear. When $V_B = V_e$ (the equivalence point) $V_B \times 10^{-pH} = 0$ (see equation (9.11). Therefore, finding the x-intercept of equation (9.13). V_e can also be deduced from:

$$V_e = -\frac{b}{m}$$

where 'b' and 'm' are the y-intercept and slope of the regression line, respectively.

Of course, $V_B \times 10^{-pH}$ never actually reaches zero and this is why an extrapolation is used. As V_B approaches V_e the Gran Plot will show some curvature. Curvature occurs because the approximations that:

1) one mole of NaOH gives one mole of A^-;

2) the HA is undissociated;

3) $\gamma_{HA} / \gamma_{A^-}$ is constant

begin to break down.

Pre-lab Calculations

Using the data posted in the Excel sheet on Blackboard:

1. Calculate the concentration of the potassium hydrogen phthalate.

2. Calculate the concentration of the NaOH. Report a percent relative standard deviation.

3. Construct a graph of pH versus volume of added base.

4. Using the data between ~0.9V_e and V_e, construct a Gran Plot and use it to determine the unknown concentration of the acetic acid.

Procedure

Prior to performing this experiment you should review weight by difference, preparing a standard solution, using a transfer pipette, performing a titration and using the Seven Easy pH meter.

Part I: Preparation of 0.030 mol L^{-1} NaOH

1. Boil (for ~5 min.) roughly 500 mL of deionized water. Boiling the water expels the CO_2 which will react with NaOH to form carbonates.

2. Using a clean graduated cylinder obtain approximately 1.5 mL of 10 mol L^{-1} NaOH. Be very careful. Concentrated NaOH is very corrosive and damaging to skin and eyes. If you get any on your skin or clothing, immediately flush with copious quantities of cold water. In the event of eye exposure, flush the eye at the eye wash station for 30 minutes. Seek medical attention.

3. Pour the 1.5 mL of 10 mol L^{-1} NaOH into a clean 500 mL polyethylene bottle. Immediately rinse the graduated cylinder with plenty of tap water followed by plenty of deionized water. If NaOH remains in the graduated cylinder, the cylinder will be damaged.

4. Fill the bottle completely with the boiled, deionized water. You do not need to measure the amount of water added.

5. Cap the bottle tightly and mix the solution thoroughly by inverting the bottle 20 - 25 times.

Part II: Preparation of a Standard Solution of Potassium Hydrogen Phthalate (KHP)

1. Using the weight-by-difference technique transfer an accurately known mass (~1.5 g) of dried and cooled KHP to a 250 mL beaker.

2. Dissolve the KHP in ~125 mL of deionized water in a 250 mL beaker and quantitatively transfer it to a 250.0 mL volumetric flask. Dilute the solution to the mark and invert the flask 20 - 25 times. Refer to the introductory pages on preparing a standard solution.

Part III: Standardization of NaOH Solution

1. Rinse the burette three times with deionized water followed by three times with NaOH.

2. Using a properly rinsed pipette, transfer 25.00 mL of the KHP solution to an Erlenmeyer flask. Add 2 - 3 drops of phenolphthalein indicator.

3. Titrate the KHP aliquot with the NaOH. Record all volumes to 0.01 mL (i.e., 1/10 of a division). Repeat the titrations until you have three acceptable data points (relative standard deviation less than 0.3%).

Part IV: Determination of the Concentration of Acetic Acid

1. Calibrate the pH meter (three point calibration with pH 4, 7 and 10 buffers).

2. Pipette 100.0 mL of the unknown acetic acid into a 250.0 mL beaker. Place a magnetic stir bar in the beaker and place the beaker on a stirring plate.

3. Immerse the pH probe and start the stirrer. Ensure the stirring bar does not strike the probe.

After a minute turn off the stirrer and record the initial pH.

4. Add 1 drop of phenolphthalein and begin titrating the acetic acid with the standardized NaOH. Initially, add NaOH in 2 mL aliquots. Allow the solution to mix thoroughly and then record the pH. Turn off the stirrer before recording the pH. Continue adding NaOH in 2 mL aliquots until the pH starts to change rapidly with an addition. Reduce the additions to 0.5 mL and when the pH starts to change rapidly again, add NaOH drop-wise until the phenolphthalein changes colour. The objective is to get plenty of data points before the equivalence point. Note the volume at which the phenolphthalein changes from colourless to pink. Record the volume and pH after <u>every</u> addition of base.

5. Once the end point has been reached, add the NaOH in 1 mL quantities (and record the pH each time) until an additional 5 mL of NaOH has been added.

Results and Calculations

1. Calculate the concentration of the potassium hydrogen phthalate.

2. Calculate the concentration of the NaOH. Report a percent relative standard deviation.

3. For part IV construct a graph of pH versus volume of added base.

4. Construct a Gran Plot and use it to determine the unknown concentration of the acetic acid.