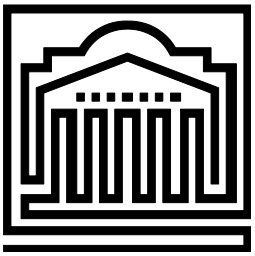
CHM 2353
Descriptive Inorganic Chemistry Laboratory

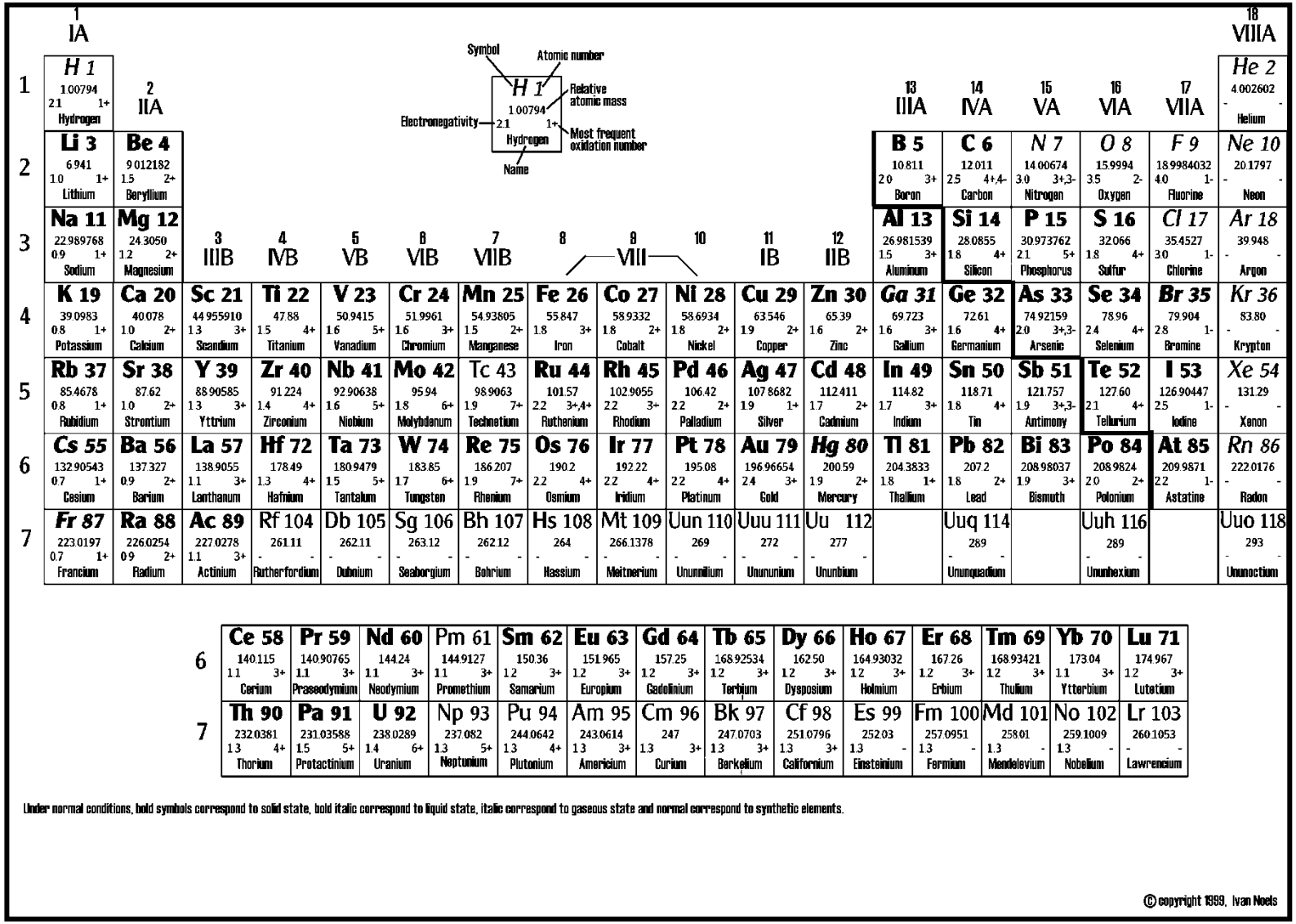
Université d'Ottawa



University of Ottawa

Laboratory Manual

Fall 2017



Under normal conditions, bold symbols correspond to solid state, bold italic correspond to liquid state, italic correspond to gaseous state and normal correspond to synthetic elements.

IMPORTANT NOTICE TO ALL STUDENTS

Safety glasses and lab coats are mandatory in all chemistry laboratories.

All students must purchase this safety equipment.

No student will be allowed to work in the laboratory without glasses and lab coat.

CHM 2353
Fall 2017

Descriptive Inorganic Chemistry

Course Instructors

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CHM 2753 Dr Eva Hemmer	DIO 118 Tel. 562-5800	ext 1987

Laboratory Instructor

CHM 2353 Dr Deryn Fogg	DIO 301 Tel. 562-5800	ext 6057
CHM 2753 Dr Eva Hemmer	DIO 118 Tel. 562-5800	ext 1987

Wednesday 18:30 - 21:30 Marion 301

Schedule of Experiments

Lab 1	**Lab 2**		
Sept. 13	Sept. 20	Exp. 0	Check-in and Volumetric Lab Techniques (**mandatory**) .....p. 6
Sept. 13	Sept. 20	Exp. I	Is solubility periodic? ..... p. 11
Sept. 27	Oct. 04	Exp. II	Spectrophotometric Determination of Nitrite and Nitrate ..... p. 14
Oct. 11	Oct. 18	Exp. III	The Copper Cycle ..... p. 21
Oct. 25			**Study Break**
Nov. 01	Nov. 08	Exp. IV	Synthetic Inorganic Chemistry: From coke can to alum p24
Nov. 15	Nov. 22	Exp. V	Analysis of Organic Nitrogen in Water by Kjeldahl Distillation..... p.27

Check Out

Organization of the Laboratory and Instructions to Students

The chemistry laboratory contains potential hazards which must be treated with respect and understanding. You are expected to arrive punctually, listen to safety instructions from the instructor and demonstrators, and proceed with your experiment with care. ***Safety glasses and a lab coat are required at all times.*** Failure to comply with the safety glasses rule may result in immediate expulsion from the laboratory.

You are expected to read the experimental procedure before coming to the lab. Pay particular attention to the chemicals you will be using, the experimental protocol and any safety precautions.

Many experiments require shared equipment. You may be charged for equipment assigned to you that is returned broken, damaged or dirty.

Attendance is compulsory at all laboratory sessions. If you have missed or will miss a laboratory session, contact the instructor as soon as possible. Just as for a missed exam, a doctor's note is required. **There is no provision for make-up experiments**. Students who miss a laboratory experiment without a valid excuse will receive a grade of zero for that experiment. Students who miss more than two experiments (even for valid reasons) will not receive a grade for the course.
The lab is worth 25 % of the total course grade.

The marking scheme for the laboratory portion of the course is as follows:

Formal laboratory reports	56 % (14/25)
Prelab preparation (in lab book)	20 % (5/25)
Laboratory technique/etiquette/lab book	16 % (4/25)
Accuracy	8 % (2/25)

Format for lab reports will be graded out of 20.
Reports must be handed according to the schedule below.

Experiment	Group 1			Group 2		
	Date Performed	Date Due	MARKED report returned	Date Performed	Date Due	MARKED report: returned
1	Sept. 13	Sept. 20	**Sept. 27**	Sept. 20	Sept. 27	**Oct. 4**
2	Sept. 27	Oct. 11	Nov. 1	Oct. 4	Oct. 18	Nov. 8
3	Oct. 11	Nov. 1	Nov. 15	Oct. 18	Nov. 8	Nov. 22
4	Nov. 1	Nov. 15	Nov. 29	Nov. 8	Nov. 22	Dec. 6
5	Nov. 15	Nov. 29	Dec. 13	Nov. 22	Dec. 6	Dec. 13

The penalty for late lab reports (without a valid medical excuse) **is 10% per day**. A report that is handed after the next lab period will not be graded.

Lab reports must be written in ink (or typed), and should be legible and brief. Your demonstrator has been instructed not to give a grade for a report that she or he cannot read!

Each report should contain the headings outlined below. Marks associated for each section may vary from report to report (obviously some experiments are more calculation intensive than others); but will ROUGHLY

Title Page or Report Header. (Marked deduction if missing, no marks allocated if complete: -5 % of report mark if any information missing)
This should include the title of the experiment, the date, your complete name and student number, your partner's name (**do not** include their student number) and your TA's name.
Experimental objectives. (5 % of report mark) State clearly in one or two sentences what the experiment is designed to demonstrate or measure.
Procedure. (5 to 10 % of report marks) Describe briefly (in a few sentences) the procedure. Do not rewrite the lab manual instructions – you may reference the lab manual rather than rewrite it. HOWEVER you should include information such as the amount of time a solution was actually boiled, how much acid was added and its **actual** concentration, etc.
Results/Observations and Discussion. (45 to 55 % of report mark) **A photocopy of signed raw data is to be attached to your report.**
This section (as all others) is to be written in sentences and paragraphs (not point form). It **MUST** include text as well as tables and graphs including a paragraph describing all visual observations (colour change, phase change, gas formation etc.) While it is a good practice to organize results into a table/graph wherever possible, the text must refer to the tables and graphs. All tables must have complete captions (located above the table) describing fully what data is presented in the table. If calculations are involved, give a sample calculation showing clearly how the calculated number was obtained. Always give the appropriate number of significant figures, and the units. Data may be presented graphically. All figures must have a complete caption (located below the figure) and should be referenced in the text. Figure and Table captions should be sufficiently detailed that the reader does not need to read the text to know what is shown.
The discussion, including interpretation of results, is the **most** important part of the report. Describe what the results mean, and include balanced chemical equations where they are appropriate. You should present some perspective on the numbers you obtain; theoretical values (such as solubility, K_{sp}, alkalinity, hardness etc.) should be referenced; do experimental values make sense - if yes why do you draw this conclusion, if not, why? What are sources of error? What else could you have done? How could the experiment have been improved? This may require outside research and should be referenced appropriately.
Calculations. (5 to 15 % of report mark) At least one example of each kind of calculation must be included using appropriate significant figures. Errors (relative or absolute) are required wherever possible.
Conclusion. **(Mark deduction if missing, no marks allocated if complete: -5 % if document does not include a quality conclusion)**
Conclusion should reflect the objectives of the experiment, and include relevant results (including sample numbers).
References. (5 % of report mark) You are expected to use scientific literature (i.e. journals), chemistry textbooks, valid internet sites to support your analysis and discussion and should include any sources you accessed – these should be presented in accepted an ACS style. If your only reference is the lab manual, your score for this section will be 0.
Questions. (20 % of report mark) Answer the questions given at the end of each experimental procedure, and any additional questions assigned by your demonstrator.

Lab books

All lab entries must be made legibly and in **INK** – this includes both prelab and all observations recorded during the lab period. Pages are not to be removed from the book. No "white out" is to be used. All entries must be dated. Your lab book is a record of the work you

complete and while it should be organized and legible it is not a work of art! **A lab technique/book grade of 0 will be assigned to students using pencil or loose papers!**

You may use partially empty books from previous courses. Any changes to procedure must be noted in the lab book. All actual volumes/masses/concentrations must be recorded. You must ensure that your demonstrator initials your data and observations before leaving the lab.

A photocopy of the signed raw data is to be attached to your formal laboratory reports.

Prelab preparation (in lab book)

Your prelab should include

1) A comprehensive flow chart or summary of the experimental procedure required for completion of the lab (see Appendix A1). This summary must highlight reagents (including volumes, masses and concentrations needed) and equipment required to complete the experiment. Your demonstrator will check that the prelab is completed prior to beginning of the lab, and if it not completed in a satisfactory manner, you will be assessed a zero for that lab.

Whenever possible your prelab should include expected outcomes (i.e. what do you expect to happen). For example, if your exercise is the evaluation of pH of various water samples (distilled water saturated with CO_2, distilled water saturated with $CaCO_3$, tap water) your prelab would include a prediction that the CO_2 saturated water should be moderately acidic, etc.

2) All hazardous materials and safety precautions required should be clearly noted.

3) A plan for disposal of chemical waste generated by the experiment should be included (i.e. solutions collected in organic waste containers, halogenated organic waste containers, disposed of in sink with water, etc.)

Laboratory technique & etiquette

The assessment of laboratory technique and etiquette will be based on the following criteria:

- arriving at the lab punctually and prepared
- recording observations and results legibly in a hardcover lab notebook
- finishing the experiment within the allotted time
- leaving the equipment, work bench and shared areas clean at the end of the lab period
- consistent use of safety glasses and other safe lab practices
- competence in the laboratory
- professional conduct

Accuracy

Several of the laboratory exercises (labs 1, 2, and 5) have an analytical accuracy component.

Experiment 0

Check in and Volumetric Lab Techniques

Part A. The burette

A burette is a precisely bored glass tube with graduations enabling you to measure the volume of liquid delivered. This is done by reading the level before and after draining liquid from the burette. The burette is first rinsed then filled with the desired solution, Figure 0-1, and the initial volume is read. ***Do not attempt to set the initial reading at 0.00 or 1.00 or any other specific reading.***

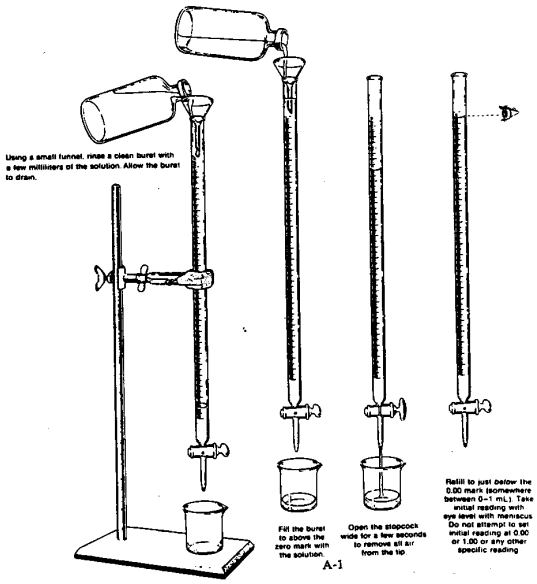


Figure 0-1. Filling the burette

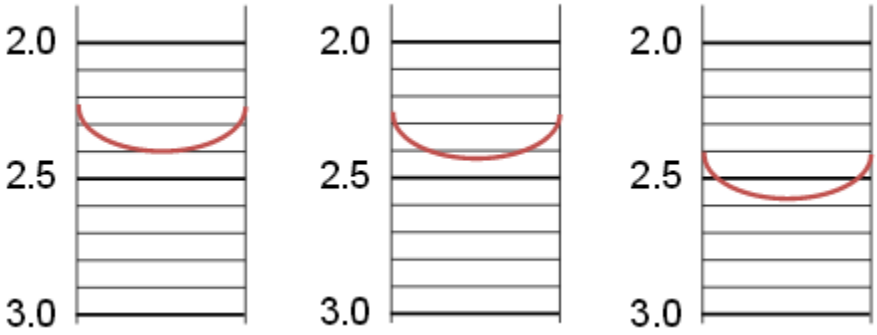
Figure 0-2. Air bubbles in the burette stopcock

If the burette is clean, the solution will sheet from the inside walls of the burette as it drains. A dirty burette will leave droplets of solution on its walls as the volume is lowered. This decreases the accuracy of the titration. Clean a dirty burette carefully with a burette brush and soapy water.

The stopcock of the burette should not leak. A leak is indicated by droplets that form at the burette tip while the stopcock is closed. If the stopcock leaks, attempt to tighten it by gently turning the Teflon nut opposite the stopcock key.

One of the most common errors in using a burette is caused by failure to expel the bubble of air that often forms directly beneath the stopcock, Figure 0-2. If an air bubble is present at the start of the titration, it may fill in during the titration, causing an error in the volume of liquid delivered from the burette. Usually the bubble can be dislodged (before beginning the titration, never during) by draining the burette for a few seconds with the stopcock wide open.

To read the volume on the burette, study Figure 0-3. Always read the bottom of the meniscus. Your eye must be at the same height as the meniscus to minimize parallax error. It is useful to use a piece of black tape on a white card as a background for locating the precise position of the meniscus. Bring the card up slowly from beneath the meniscus until you see it sharpen. Align the bottom of the black tape with the bottom of the meniscus and read the position on the burette. The markings on the burette normally have a thickness, which is not negligible in comparison with the distance between markings. The thickness of the markings corresponds to about 0.02 mL. You must select one part of the marking to be called zero. For example, say that the liquid level is at the mark when the bottom of the meniscus just touches the top of the mark on the glass. Call this level zero. When the level is at the bottom of the mark, the reading is 0.02 mL higher. Similarly, when the level is one line thickness higher than the mark, the reading is 0.02 mL lower. The burette can and should be read with an accuracy of 0.01 mL.



Reading: 2.40 2.42 2.58

Figure 0-3. Reading the volume on a burette

To deliver solution from the burette, open the stopcock. The recommended technique for manipulation of the burette stopcock is shown in Figure 0-4. Most left-handed students will prefer to manipulate it with the right hand (A), whereas most right-handed students will prefer to manipulate it with the left hand (B). The solution should be delivered into an Erlenmeyer flask (not a beaker) held in the opposite hand and swirled continuously. For titrations, a piece of white paper beneath the flask makes the endpoint easier to detect. Near the endpoint, solution from the burette must be delivered one drop at a time. Drops hanging from the burette tip are already counted as volume delivered. They can be added to the Erlenmeyer flask by gently touching the burette tip to the side of the flask and washing drop down into the flask with distilled water from a squirt bottle.

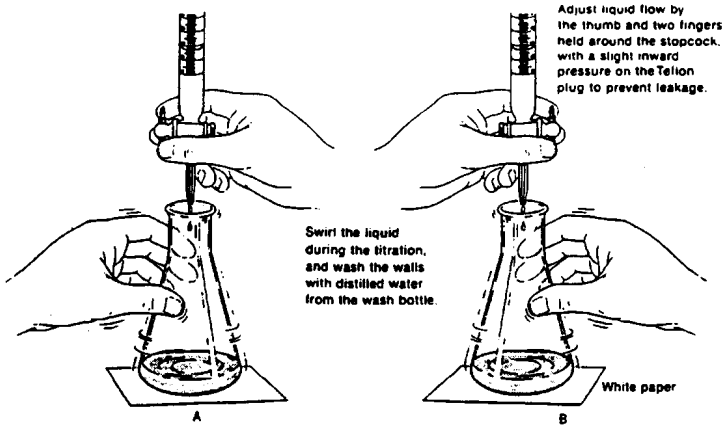


Figure 0-4. Delivering solution from the burette

The first time a titration is performed, the delivery of solution must be done slowly to avoid overshooting the endpoint. In subsequent titrations, you may quickly deliver a volume equal to one mL less than the titration volume then proceed drop-by-drop to the endpoint.

Part B. Volumetric flasks

A volumetric flask is calibrated to contain a particular volume of water at 20°C when the bottom of the meniscus is adjusted to the center of the line marked at the neck of the flask. A volumetric flask is used to prepare a solution of known volume. Most commonly, the reagent is added to the flask and then diluted to the mark. The final adjustment to the mark should be done with a Pasteur pipette, not with a beaker or a squirt bottle. After dilution insert the stopper and invert several times to assure complete mixing.

The final concentration of the solution takes into account the precise dilution. For example, a 10 mL aliquot of a stock solution containing 100 ppm chloride, when diluted in a 50 mL volumetric flask, will have a concentration of 100 x (10/50) = 20 ppm. The factor 10/50 is called the *dilution factor*.

Part C. Volumetric pipettes

Volumetric pipettes (as opposed to Pasteur pipettes) are used to deliver known volumes of liquid. The most common types are the transfer pipette and the measuring pipette. The transfer pipette is calibrated to deliver one fixed volume. The last drop of liquid does not drain out of the pipette; it is meant to be left in the pipette and ***should not be blown out.*** However, the tip must be intact for the calibration to be valid. Never use a pipette with a chipped tip. The measuring pipette is calibrated to deliver a variable volume, as indicated by the difference between the volumes indicated before and after delivery. Again, the last drop must not be blown out.

The use of the volumetric pipette is shown in Figure 0-5. Use a red plastic pipette pump to suck liquid up past the calibration mark. Touch the tip of the pipette to the side of a beaker and use the release valve to slowly drain the liquid until the bottom of the meniscus reaches the center of the calibration mark. The pipette must touch the side of the beaker during draining so that extra liquid is not hanging from the tip of the pipette when the meniscus

reaches the mark. Transfer the pipette to the receiving flask and drain it in a nearly vertical position, while holding the tip to the wall of the flask.

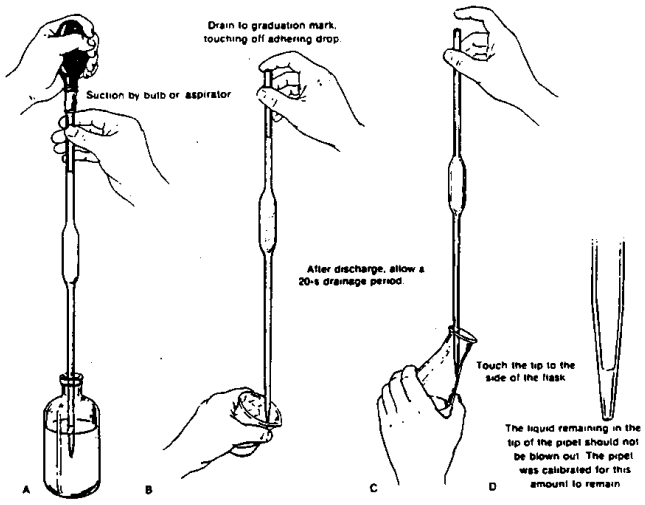


Figure 0-5. Using a volumetric pipette

Alternatively, suck the liquid up past the calibration mark with the pipette pump, quickly remove the pump and replace it with your index finger (see figure below). Touch the tip of the pipette to the side of a beaker and drain the liquid out until the bottom of the meniscus reaches the center of the calibration mark. Transfer the pipette to the receiving flask and drain it in a nearly vertical position, while holding the tip to the wall of the flask

Common sources of error during pipetting are (1) cross-contamination of solutions; and (2) aspirating solution into the rubber pipette bulb. Make sure the pipette bulb is dry during the entire operation. You should never put a volumetric pipette directly into a stock solution bottle. Always transfer a slightly larger volume than you need into a small flask and pipette from this flask. Wipe the tip of the pipette to remove liquid from the outside of the pipette. Begin aspirating with the pipette bulb before immersing the tip of the pipette into the solution so that any drops remaining in the pipette do not contact the stock solution. Pull a few mL of solution into the pipette, turn the pipette to a horizontal position and rotate so that the solution contacts all the interior surfaces of the pipette below the calibration mark. Drain the solution out of the pipette and repeat this operation twice more.

Check in

Obtain your locker assignment from your demonstrator. Verify the equipment in your locker against the list at the end of this manual. Replace any equipment that is broken or chipped.

Experimental procedure

Remember you must record your observations <u>in pen</u> and <u>in a lab notebook.</u> In this experiment, each student will **<u>work independently</u>**.

Part A. The burette

Place a piece of black tape on an index card to help you read the position of the meniscus on the burette. Fill a burette with distilled water and read the initial volume to the nearest 0.01 mL (*e.g.*, 1.03 mL). Ask your neighbor to read the volume and compare your answers.

Weigh a small beaker to the nearest 0.1 mg on the analytical balance. Deliver about 20 mL of distilled water from the burette and read the final volume. Reweigh the beaker.

Repeat the operation three times.

Part B. The volumetric pipette

Weigh a small beaker to the nearest 0.1 mg on the analytical balance. Deliver precisely 20 mL of distilled water from a volumetric pipette. Reweigh the beaker. Repeat the operation three times.

Draw a small volume of the purple permanganate solution into your pipette. Let the permanganate drain into a beaker and push the remaining drops of permanganate out of the pipette with the pipette bulb. Rinse the pipette by drawing a few mL of distilled water into the pipette and rotating it horizontally. (The distilled water in the beaker should not become purple during this operation!) Repeat the rinsing until you can no longer detect the purple color.

No report required.

Experiment I

An introduction to periodic trends or Is solubility periodic?

Descriptive Inorganic Chemistry is a survey course of the chemistry of main group elements and relevant background theory pertaining to this chemistry. It makes use of the elegance and some might say, brilliance, of the periodic table which has been constructed so that elements exhibiting similar chemical behaviours are grouped together. In this laboratory exercise the solubility of ionic hydroxide salts of Group II metal cations will be investigated to draw attention to periodic trends.

Ionic solids consist of repeating arrays of positively and negatively charged ions. The attraction between ions is electrostatic (or Coulombic) in nature and the strength of these attractions reflects ionic charge and size (interestingly, these, too, show periodic trends). Many ionic compounds dissolve in water forming conductive solutions (e.g. salt, NaCl) while others dissolve to a significantly lesser degree (e.g. limestone, $CaCO_3$). The extent to which ionic compounds dissolve is the property known as solubility. The solubility of simple salts was introduced both qualitatively (solubility rules) and quantitatively (equilibrium constants, K_{sp}) in General Chemistry.

Consider the reaction shown below describing the dissolution of a sparingly soluble group II metal hydroxide (M represents the group II metal):

$$M(OH)_2(s) \rightleftarrows M^{2+}(aq) + 2OH^-(aq) \qquad (1.1)$$

As always the arrow, $\rightleftarrows$, indicates a reversible process in which both solid salt and dissolved ions are present. The position of the equilibrium, and of course the numerical value of K_{sp}, is governed by thermodynamics (i.e. enthalpy and entropy of the dissolution process).

If the system is set up so that the maximum amount of salt can dissolve (as noted by excess solid salt settled at the bottom of the reaction vessel) then it will, overtime, reach equilibrium. At this point, the concentrations of the ions in solution will no longer change (although reaction (1.1) above continues to occur; remember equilibrium is dynamic).

Equilibrium constants, including the solubility product, K_{sp}, are determined by the ratio of activities of products to reactants. Often, concentrations are used to approximate activities and in the above case:

$$K_{sp} = [M^{2+}][OH^-]^2 \qquad (1.2)$$

Clearly, evaluation of the concentration of one of the ionic species can be used to determine the solubility product (or, more accurately, to approximate the solubility product). In this case, a titration of the saturated salt solution against a known concentration dilute strong acid solution is used to determine the concentration of OH^-.

$$H_3O^+(aq) + OH^-(aq) \rightarrow 2\ H_2O \qquad (1.3)$$

Phenolphthalein is used to determine the endpoint and thus estimate the equivalence point of the titrations.

Procedure:
Saturated solutions of $Mg(OH)_2$, $Ca(OH)_2$ and $Sr(OH)_2$ have been prepared. Standard solution of 0.0020 M HCl is also available.

Reminder: always record volumes on the burette to the nearest **0.01 mL**. Near the endpoint, add the titrant drop by drop and swirl the solution constantly. The end of the titration is the first drop of titrant which causes a perceptible colour change. If the endpoint of the titration is not clear, repeat the experiment using more indicator.

Wash all glassware prior to use (this includes, but is not limited to the burette, pipettes, funnels, beakers, Erlenmeyer flasks, graduate cylinders).

Rinse the burette with several small (1 or 2 ml) aliquots of standard HCl solution. Fill the burette with the standard HCl solution (ca. 0.002 M) and ensure that no air bubbles are trapped in the stopcock (also, don't forget to record its exact concentration in your lab book).

Rinse a volumetric pipette with several small aliquots of saturated $Mg(OH)_2$ solution, then use the pipette to transfer 50.0 mL of saturated $Mg(OH)_2$ solution to a clean Erlenmeyer flask. Add several drops of phenolphthalein indicator and record the colour (or lack thereof). Titrate with standard dilute HCl as required. Repeat the titration at twice (for a total of three $Mg(OH)_2$ samples).

Repeat procedure beginning with 1.0 ml of $Ca(OH)_2$ transferred to a clean Erlenmeyer flask; dilute with 50 ml distilled water prior to titration. (3 titrations required).

Repeat above procedure beginning with 0.5 ml of $Sr(OH)_2$; dilute with 50 ml of distilled water prior to titration.

Report:

Results should be summarized in a Table; the initial volume, final volume, volume added and moles of acid required to reach the endpoint, as well as concentration of OH^-, K_{sp} and solubility, s, in mg L^{-1} for **each** trial should be included. Report also the average concentration of OH^-, K_{sp} and the solubility of each salt. Show a sample of each calculation.

Compare experimental results to values available in literature – be sure to reference the literature values. Are values in agreement with literature?

Determine a trend in their experimental results and connect the trend to periodicity. Are trends in agreement with literature? Explain (based on chemical properties) observed trend / lack of trend.

Discuss why experimental results deviate from literature values.

Questions:
1. Would the solubility of $Mg(OH)_2$ in 0.01 M hydrochloric acid be greater than, or less than that in pure distilled water? Explain. Would a similar result be observed in phosphoric acid ?

2. What volume of a saturated solution of barium hydroxide would you suggest using (keeping in mind the glassware you have available) if you wanted to determine solubility of $Ba(OH)_2$ using a 0.002 M solution of HCl. Explain your answer.

3. In the experiment to determine the solubility of alkaline earth hydroxides using hydrochloric acid solution:

a. What is the balanced net ionic reaction occurring during EVERY titration in this experiment?

b. What is the analyte in the titration?

c. What is the titrant solution in the titration?

d. What else do you need to add to the analyte before completing the titration?

e. How do you know when the titration is over?

f. Is there a difference between endpoint and equivalence points? Explain.

Experiment II

Spectrophotometric Determination of Nitrite and Nitrate

The nitrogen cycle

The environmental chemistry of nitrogen is complex because the element exists in many different oxidation states, which can be interconverted by living organisms. In aquatic systems, the nitrogen compounds of most importance to water quality are ammonia (NH_3), nitrite (NO_2^-) and nitrate (NO_3^-).

The nitrogen in these compounds originates in the atmosphere, which is essentially an inexhaustible reservoir. N_2 is constantly removed from the air, or *fixed*, by the action of lightning and certain bacteria. During electrical storms, N_2 is oxidized to NO_2, eq 2.1, which is further oxidized by the hydroxyl radical to nitric acid, eq 2.2.

$$N_2 + 2\,O_2 \rightleftarrows 2\,NO_2 \qquad (2.1)$$

$$NO_2 + HO \rightleftarrows HNO_3 \qquad (2.2)$$

Nitric acid is carried to the ground in the rain where it serves as the source of nitrogen for protein synthesis by growing plants and, subsequently, animals. N_2 can also be converted directly to proteins by nitrogen-fixing bacteria. Nitrate is produced *anthropogenically* as a commercial fertilizer by oxidation of ammonia in the presence of a catalyst.

The nitrogen found in proteins and urea is called *organic nitrogen*. Once an organism dies, its organic nitrogen is recycled and reused by other plants. First, it is converted to ammonia by heterotrophic bacteria, under both aerobic and anaerobic conditions. Ammonia is oxidized by autotrophic nitrifying bacteria. *Nitrosomonas* bacteria, also known as nitrite-formers, convert ammonia to nitrite, eq 2.3.

$$2\,NH_3 + 3\,O_2 \rightarrow 2\,NO_2^- + 2\,H^+ + 2\,H_2O \qquad (2.3)$$

Nitrite is then oxidized to nitrate, eq 2.4, by *Nitrobacter* bacteria, which are called nitrate-formers.

$$2\,NO_2^- + O_2 \rightarrow 2\,NO_3^- \qquad (2.4)$$

The form of nitrogen that is directly assimilated by plants is nitrate (hence the use of nitrate salts as fertilizers). Under anaerobic conditions, nitrate can be reduced by bacteria in a process called *denitrification*, which converts nitrate to nitrite and then to N_2. A small amount of nitrate is partially reduced to the greenhouse gas N_2O.

Health concerns about nitrite and nitrate in drinking water

Nitrate salts are very soluble. In addition to being assimilated by plants, they dissolve in the water percolating through the soil. This results in high ground water concentrations of nitrate in agricultural areas. Nitrate and nitrite can also enter the drinking water supply as

a result of the presence of nitrifying bacteria in water treatment plants.

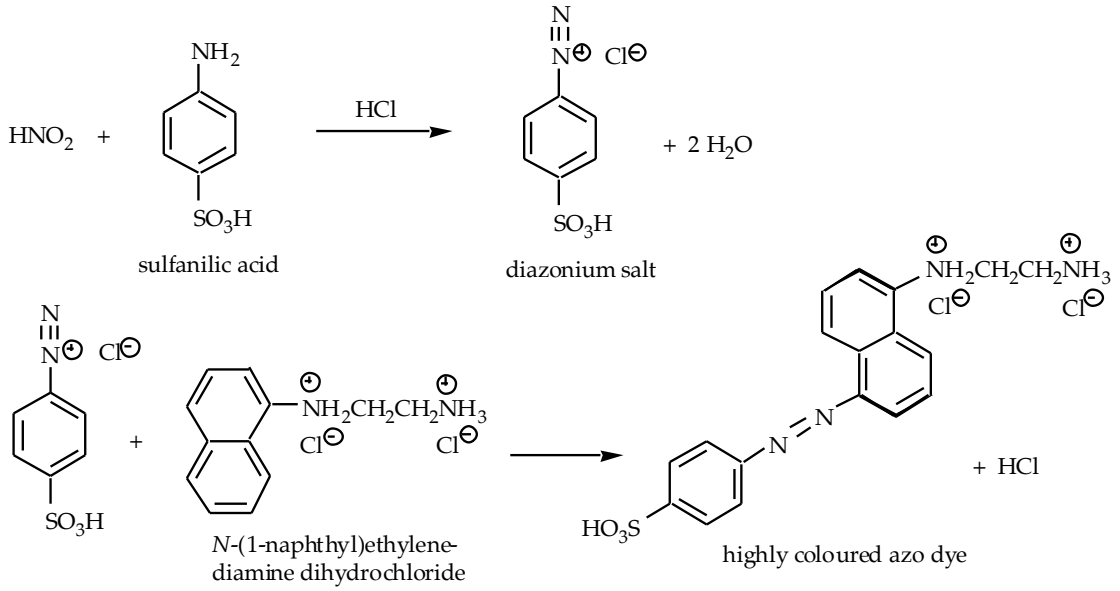
Drinking water with high nitrate content is linked to *methemoglobinemia* (blue baby syndrome). The direct cause of methemoglobinemia is the nitrite ion. It can be formed when nitrate is reduced by *E. coli* bacteria which colonize the upper intestine of infants. (This reaction does not occur in adults, whose intestinal tracts are more acidic.) Nitrite oxidizes the oxygen-carrying ferrous ion (Fe (II)) of hemoglobin. The resulting ferric (Fe (III)) hemoglobin (called methemoglobin) cannot transport oxygen.

In highly acidic adult stomachs, directly ingested nitrite reacts with secondary amines (RR´NH) to form nitrosamines (RR´NNO), some of which are known to be carcinogenic. The toxicology of nitrite is therefore the subject of much current research. The Ontario Drinking Water Objectives formulated by the Ministry of Environment and Energy require that the nitrate-N concentration not exceed 10 mg/L in public water supplies, while nitrite-N must not exceed 1.0 mg/L. (For the definitions of nitrate-N and nitrite-N concentrations, see the Note about concentrations at the end of this laboratory protocol).

Nitrate and nitrite analyses can be used to detect pollution of water by sewage. Raw sewage contains pathogenic bacteria, but the direct detection of their presence is a lengthy process requiring several days for culturing. In contrast, analyses for nitrite and nitrate are quick and convenient. These forms of nitrogen arise from the denaturing of proteins and the oxidation of the ammonia by bacteria. Such indirect analyses of water quality have now been superceded by tests for less harmful coliform bacteria (such as *E. coli*) which usually accompany the more dangerous strains but are much easier to detect. Analyses for nitrate and nitrate are now only performed to detect contamination by agricultural pollution.

Analysis of nitrite and nitrate

Unless the concentration of dissolved oxygen is very low, nitrite-N seldom appears in concentrations greater than 1 mg/L, even in water treatment plant effluents. Its concentration in surface and groundwaters is usually << 0.1 mg/L, and it is efficiently oxidized by Cl_2 to nitrate during the purification of drinking water. For these reasons, a very sensitive method is required for its measurement. We will use a diazotization reaction, which produces a highly coloured azo dye. Under acidic conditions, the nitrite ion is protonated to form nitrous acid, which reacts with the amino substituent of sulfanilic acid to form a diazonium salt. The latter combines with *N*-(1-napthyl)-ethylenediamine dihydrochloride to form a intensely coloured azo dye. The intensity of the colour is directly proportional to the amount of nitrite in the sample.



In order to use this procedure to analyse for nitrate, the nitrate must be partially reduced by zinc to nitrite. The amount of zinc and its time in contact with the nitrate solution must be carefully controlled. Since nitrite originally present in the sample will also be detected by the diazotization reaction, the resulting colour intensity represents the contributions of both nitrate and nitrite. In order to determine the nitrate concentration alone, a separate analysis of nitrite must be performed without the Zn reduction step. The nitrate concentration is then calculated as the difference between the two analyses.

Experimental procedure

At the beginning of the lab period, the demonstrator will assign each student a sample for analysis. Note the sample number in your lab book. Students work in pairs to prepare the standard solutions but each student should **independently** determine the concentration of an unknown.

Note: The order of addition of reagents is important; follow the order given. Before coming to the laboratory, calculate the volumes of stock solutions required and prepare a table in your lab book to fill in during the experiment.

Part A. Nitrite analysis

Prepare standard solutions of 0, 0.2, 0.4, 0.6 and 1.0 ppm nitrite by pipetting the appropriate amount of stock nitrite solution (ca. 20 ppm) into 100 mL volumetric flasks. Measure with a disposable plastic pipette and add to each flask 2 mL dilute HCl and 2 mL sulfanilic acid. Add 2 mL *N*-(1-naphthyl)ethylenediamine dihydrochloride solution and 2 mL 2 M sodium acetate. Dilute to the mark with distilled water. Mix well by stoppering the flasks and inverting several times. Let the standard solutions stand for 5 minutes.

In each of two 100 mL volumetric flasks, pipette a 25 mL aliquot of your unknown sample and follow the nitrite analysis procedure given above.

Determine the optimal wavelength (LabQuest2 and Spectrovis).
In this part of the experiment you will determine the optimum wavelength at which to measure all future samples by measuring and analyzing the visible spectra for you 1.0 ppm sample.

1. Turn the LabQuest 2 on.
2. Connect the SpectroVis to the LabQuest 2 via the USB port.
3. Fill a plastic cuvette with the blank solution (0 ppm nitrite). Wipe the outside of the cuvette with a Kimwipe.
4. To calibrate the SpectroVis, click anywhere on the red box and select **Calibrate**.
5. Place the cuvette containing blank solution in the SpectroVis and select **Finish the Calibration** and press **OK**.
6. Remove the cuvette
7. Rinse and fill the cuvette with 1 ppm standard solution. Use a Kimwipe to wipe the outside of the cuvette.
8. Insert the cuvette into the SpectroVis and ensure that Labquest 2 **Mode** is set to **Full Spectrum**
9. Start the run by clicking on the green arrow in the bottom left hand corner of LabQuest 2.
10. Once the spectrum is complete, click on the small red square (stop), select **Analyze** tab (at top of screen), select **Statistics**, and select **Abs**
11. The wavelength at which maximum and minimum absorbance will be displayed. Note the appropriate value in your lab book – you will use this for all future measurements.
12. Use the **File** tab (top left) to **Save** the run to LabQuest 2 (make sure you note the file name!).

This spectrum **MUST** be included in your formal report.

Measure absorbance of standard solutions and unknowns
Next you will measure the absorbance of each of your standard solutions and unknowns at the wavelength selected base on you results above.

1. Click on the **Sensor** icon (top left-most corner of screen)
2. Return to **Mode**, select **Events with Entry**, **Set Columns** to **1**, Name the column concentration (with appropriate units), click **OK**
3. Click on the **red square**, choose **Select Wavelength**, and enter the wavelength determined in 11 above.
4. Rinse cuvette thoroughly with test solution (start with lowest concentration). Fill cuvette and dry the outside.
5. Place cuvette in SpectroVis, click the **green arrow** (lower left) to start data acquisition. When you are satisfied with the value, click the **Keep** icon to the right of the green arrow. Enter the sample concentration.
6. Empty the cuvette, rinse with next most dilute solution, measure absorbance. Repeat.
7. Use the **File** tab (top left) to **Save** data.

You may also record the appropriate values directly in your lab notebook.

Prepare a calibration curve by plotting absorbance vs. nitrite concentration (you may use

Excel or similar graphing software; make sure your work is done independent of your lab partner). Based on the calibration curve, determine the nitrite concentration in your sample solutions.

Part B. Nitrate calibration curve

TIMING IS CRITICAL

Prepare standard solutions of 0, 5, 10, 15 and 20 ppm nitrate by pipetting the appropriate amount of stock nitrate solution (ca. 500 ppm) into 100 mL volumetric flasks. At the same time, prepare two sample solutions using 25 mL aliquots of your unknown. To each flask, add 2 mL dilute HCl and 2 mL sulfanilic acid. To ensure a reproducible contact time with the zinc, perform the next step on all the solutions as close to the same time as possible.

In the next stage of the procedure **TIMING IS CRITICAL (Why?).** Measure approximately 2 g Zn/NaCl powder for each sample and add to each flask and swirl. Let the solutions stand over Zn for five minutes.
Add 2 mL N-(1-naphthyl)ethylenediamine dihydrochloride and 2 mL 2 M sodium acetate. Dilute to mark with distilled water.

YOUR EXPERIMENT WILL NOT WORK UNLESS THE TIME OF EXPOSURE TO Zn (TIME BETWEEN ADDITION OF Zn/NaCl POWDER AND ADDITION OF THE SODIUM ACETATE) IS IDENTICAL FOR ALL SAMPLES.

Wait a further 5 minutes for colour development, then measure the absorbance of each solution, starting with the most dilute standard. Measure all absorbances at as close to the same time as possible.

Prepare a calibration curve by plotting absorbance vs. nitrate concentration.

Your unknown initially contains both nitrite and nitrate. To calculate the nitrate concentration in your sample, first subtract the absorbance due to nitrite (from Part A) Then read the nitrate concentration from the nitrate calibration curve.

To transfer data to USB key (both full spectrum data and concentration data):

1. Plug USB key into LabQuest 2
2. To save calibration curve or absorbance data to the USB key, first click on **File**, then **Open**, the click on the LabQuest2 icon, select appropriate file.
4. From the **File** menu choose **Export**
5. Tap **USB icon** (top of screen)
6. Name file (**DONE**) and choose **OK**

21. To import .txt (Tab limited file into Excel, MAC)

1. Open Excel
2. Click **File**
3. Click **Import**
4. Choose **Text file**, then click **Import**

5. Highlight file to be imported
6. Click **Get Data**
7. File is **Delimited**, Start import in first row, Click **Next**
8. Delimiter is **Tab**, Click **Next**
9. Click **Finish**

21. To import .txt (Tab limited file into Excel, PC)
    1. Open Excel
    2. Click **File**
    3. Click **Open**
    4. Choose **Text Files** (lower right hand side)
    5. Highlight file to be imported
    6. Click **Open**
    7. File is "**Delimited**", Start import in first row, Click **Next**
    8. Delimiter is **Tab**, Click **Next**
    9. Click **Finish**

Report

Show the full spectrum and indicate the wavelength used in all subsequent experiments. Present both calibration curves (remember to use appropriate labels on all axes) and lines of best fit. Report the concentration of nitrite and nitrate in your unknown, taking into account its dilution factor. Include sample calculation.

Your report should include all relevant chemical reactions, including but not limited to the formation of the coloured species, reaction of nitrate, etc.

Why is it important to measure absorbance starting with the least concentrated solution and finishing with the most concentrated solution? According to the Ontario Drinking Water Objectives, is your water sample potable? What is the absorbing species in part A ? in part B? Why is the nitrite absorbance in part A subtracted from the experimentally measure absorbance in part B prior to determining the nitrate concentration? How are nitrates/nitrites removed from drinking water ?

Questions

1. Based on your experimental data, estimate the % nitrate converted to nitrite during 5 minute exposure to zinc
2. The introduction states that nitrate is partially reduced to nitrite:
    a. What are the chemical differences between the nitrate and nitrite ions (oxidation states, bond strengths, oxidizing ability)? You may want to draw Lewis structures.
    b. What was oxidized? (include appropriate chemical reaction)
    c. Why is the time that the nitrate solution and zinc powder are in contact an important factor in this procedure?

Note about concentrations

In chemistry, the preferred unit of concentration for species in solution is mol/L. However, in the industrial world, many other units are used and you must be able to recognize what they mean and know how to convert them. Parts per million, abbreviated as ppm, is a mass ratio of 10^{-6}. For example, 1 mg of substance X dissolved in 1 kg of solvent is said to be present at the level of 1 ppm. Since water has a density close to 1 kg/L at room temperature, 1 ppm of substance X in water is equivalent to 1 mg/L. For very low concentrations, parts per billion (ppb) or parts per trillion (ppt) may be used.

When an element occurs in many different forms in the environment, as does nitrogen, it is convenient to make comparisons based on elemental masses. For example, nitrogen can exist in natural waters as ammonia, nitrite or nitrate. The safe limit for nitrate in drinking water is 45 ppm NO_3^- (formula mass of the nitrate anion is 62g $mole^{-1}$), which is equivalent to 10 ppm nitrate-N (atomic mass of nitrogen is 14 g $mole^{-1}$). 10 ppm nitrate-N means that the solution contains 10 mg of nitrogen from dissolve nitrate per liter of solution. To convert concentration from ppm to ppm nitrate-N you must first determine the concentration of nitrogen in moles L^{-1}.

For example:

$$
\begin{aligned}
45\ ppm\ NO_3^- &= 45\ mg\ of\ NO_3^-\ per\ litre\ of\ solution \\
&= \frac{45x10^{-3} g\ L^{-1}}{62g\ mole^{-1}} \\
&= 7.26x110^{-4}\ mole\ L^{-1}\ *\ 14\ g\ of\ N\ mole^{-1} \\
&= 1.02x10^{-3}\ g\ N\ mole^{-1} \\
&= 10\ mg\ N\ mole^{-1} \\
&= 10\ ppm\ Nitrate - N
\end{aligned}
$$

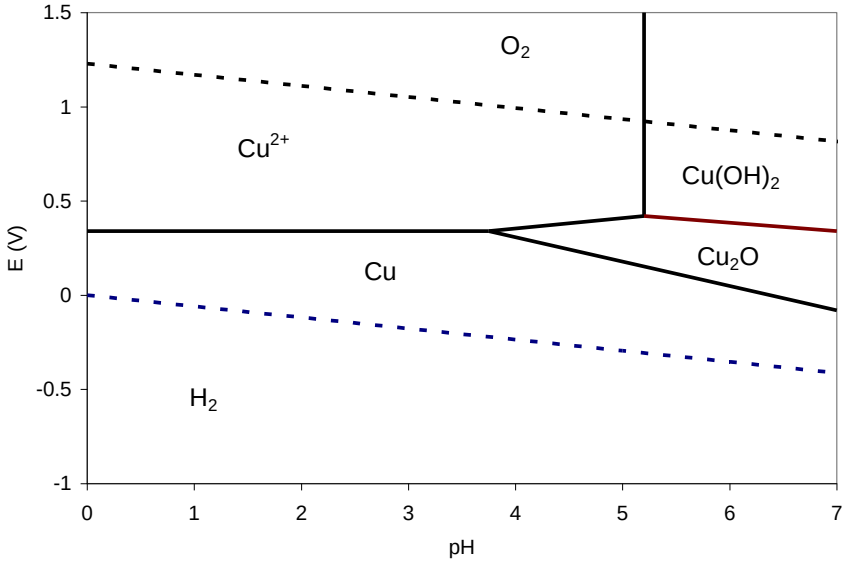
Experiment III

The Copper Cycle

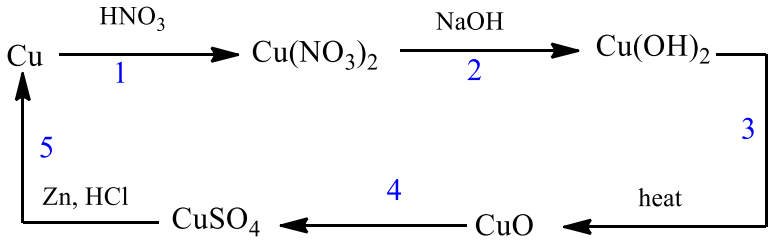
Copper, a member of group 11, is called a coinage metal (along with silver and gold) for obvious reasons. It exhibits oxidation states of +1 and +2 and, while not abundant in nature, can be extracted from numerous ores by pyrometallurgical (roasting) or hydrometallurgical (leaching in dilute acid solution) processes. The +2 oxidation state dominates aqueous copper chemistry and the characteristic blue colour of copper solutions is due to the presence of the hexaaquacopper (II) ion, $[Cu(OH_2)_6]^{2+}$. Aqueous solutions of Cu(I) are unstable with respect to disproportionation as indicated in the Latimer diagram below.

$$Cu^{2+}(aq) \xrightarrow{0.159\ V} Cu^{+}(aq) \xrightarrow{0.520\ V} Cu(s)$$

A simplified Pourbaix diagram for Cu in aqueous solution (below) illustrates the copper speciation as a function of pH.



In this laboratory exercise you will conduct a series of reactions that involve the element copper. The reaction scheme begins and ends with copper metal, and as no copper is added or removed at any time during the cycle and because each of the reactions is heavily product favoured, you should be able to quantitatively recover all the copper you start with. It is critical that you carefully observe and document all physical/chemical changes that occur during the sequence of reactions. The major reactants and products are noted in Reaction Scheme below:



Experimental Procedure

Step 1: Cu to $Cu(NO_3)_2$: Measure out a 0.5 g sample of pure copper wire (approximately 7 cm) to the nearest milligram. If it is not bright and shiny, clean it with steal wool, rinse with water, dry and re-weigh. Alternatively, the Cu wire can be clean my swirling in a small beaker containing 5 ml of 6 M HCl, then rinsed with water, dried and reweighed.

Next steps to be **performed in the fumehood**.

Place coiled wire in beaker and add 4 ml of concentrated (16 M) HNO_3. Swirl beaker gently until all copper has been completely dissolved. ***Slight*** heating may be required to complete the reaction. Note any changes in the colour of the solution and changes in state (formation of gases or solids).

Once reaction is complete, add 100 ml of distilled water.

Step 2: $Cu(NO_3)_2$ to $Cu(OH)_2$: (May be done on the bench) Add, with constant stirring, 30 ml of 3.0 M NaOH. Confirm that reaction has gone to completion by testing a drop of the solution with litmus paper (use a stirring rod to place a drop of solution on the paper; do not place the litmus paper directly in the solution). If reaction is not complete, add an additional 5 ml of NaOH and test with litmus paper. If necessary, repeat.

Note any changes in the colour of the solution and changes in state (formation of gases or solids). Note also colour of litmus paper.

Step 3: $Cu(OH)_2$ to CuO: (May be done on the bench) Heat the solution to just barely boiling, stirring continuously). If you fail to stir, the solution will bump and product will be ejected from beaker. If the expected transformation is not observed within 10 minutes of heating, add approximately 10 ml additional sodium hydroxide solution and continue to heat. When transformation to CuO is complete, heat for an additional 2-3 minutes then remove beaker from hot plate, continue stirring for several minutes and then allow to cool and settle. Note any changes in colour of solution or solids and any changes of state.

Isolate the copper oxide by vacuum filtration using a Buchner funnel. Wash the solid product with two portions of 25 ml of distilled water.

If filtrate is clear, discard. If particles appear in the filtrate, re-filter onto the same piece of filter paper until solution is clear.

Step 4: CuO to $CuSO_4$: Quantitatively transfer the solid (and filter paper) from the filtepaper to a 250 ml beaker (you may use your spray bottle to ensure a quantitative transfer of the solid). Add 15 ml of 6.0 M H_2SO_4 (***slowly, with stirring***) to dissolve the solid. Heat the mixture gently, if required.

Note any changes in colour of solution or solids and any changes of state.

Clean the filter funnel and flask and re-filter your mixture to remove the filter paper particles from the filtrate (the liquid that passes through the filter). You will want to keep the liquid. Rinse beaker with a small volume of distilled water and pass through the filter. Transfer filtered solution to a clean 250 ml beaker.

Step 5: $CuSO_4$ to Cu metal: (Back in Fumehood!). Fill an empty 250 ml beaker with water, add boiling chips and place on hot plate to boil (to be used in drying of Cu metal).

In fumehood, add all at once, 2.0 g of zinc metal powder (2 fold excess), stirring until liquid is colourless and all bubbling has ceased. If bubbles continue to form, warm the copper solution on hot plate until gas evolution stops. This step may take 10 to 15 minutes. To ensure all copper has been reduced, obtain 1 ml of concentrated ammonia and add a drop of the copper solution. Observe any color changes (unreduced Cu/ammonia turns a deep blue); if needed allow copper / zinc mixture to react longer.

Once all copper has reacted, there should be no silvery pieces of Zn remaining in the solution. If you can see unreacted zinc, add 10 ml of 6 M HCl and warm (DO NOT BOIL) the solution. Continue gently heating solution until all gas is driven off.

Decant (pour with care) the liquid from the copper into a separate beaker. Wash product with 4 ml of distilled water, allow to settle, then discard the wash water. Repeat washing two more times.

Transfer copper to a dry watch class and wash with 5ml of methanol, allow the precipitate to settle and then decant the methanol wash. Repeat.

Dry copper by placing watch glass over the beaker of boiling water. Use stirring rod to move the copper pieces around to help drive off the methanol. Do not heat copper past "dryness" or it will be oxidized. Allow copper time to cool, and determine final weight of copper recovered.

Note any changes in colour of solution or solids and any changes of state.

Report

Compare beginning and ending amounts copper and express as a percent yield for the reaction cycle.

Discussion

Propose thermochemical reactions for each step in the cycle and explain how your physical observations support the proposed reactions. Comment on You should also include appropriate references to support your proposed reactions.

Questions

1. 0.45 g of Cu is dissolved in 4.0 ml of 12 M HNO_3. What mass of solid sodium hydroxide pellets must be added to precipitate the Cu^{2+} as $Cu(OH)_2$?

2. What would be the effect of adding 300 ml of 0.3 M NaOH rather than 30 ml of 3 M NaOH in the conversion of $Cu(NO_3)_2$ to $Cu(OH)_2$? How would this change the experimental procedure for the remaining steps?

Experiment IV

Inorganic Synthesis from Coke can to Alum

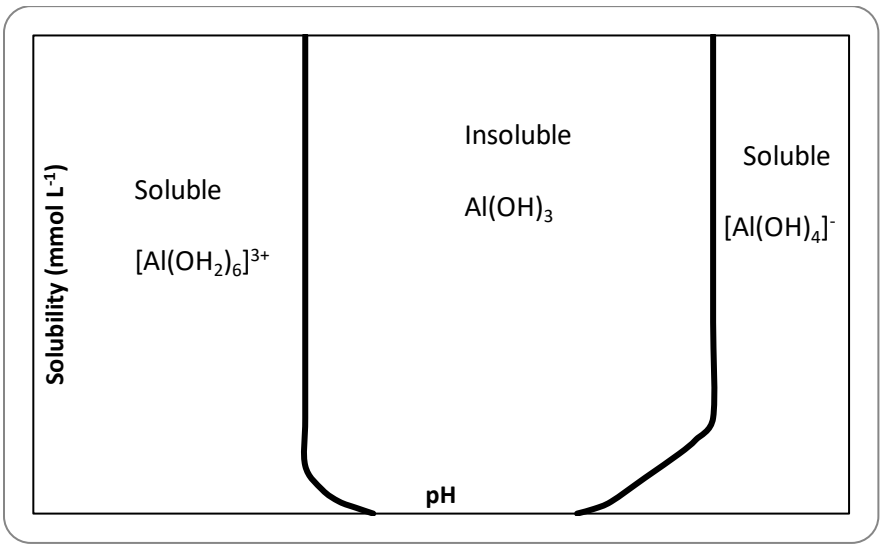
Aluminum is the most abundant metal in the Earth's crust; 7.5 % (by mass) and 6.5% (by # atoms) of the crust is aluminum, primarily in the form of clays (aluminosilicates). It is not possible to economically extract aluminum from clay, however, extraction form bauxite (hydrated aluminum oxide) formed in hot humid environments is possible. Further, the chemical properties of aluminum are interesting, including its low density (2.7 g cm^{-3}, cf. 7.9 g cm^{-3} for iron and 19.3 g cm^{-3} for gold), good thermal and electrical conductivity and reactivity in both acids and bases (amphoteric nature of aluminum and its oxides).

One family of aluminum compounds are the alums. Alums have the general form $M^{+}M^{3+}(SO_4^{2-})_2 \bullet 12H_2O$; the monopositive center is generally potassium, rubidium or ammonium and the tripositive cation is often aluminum, chromium(III) or iron(III). Alum refers to the aluminum containing compound, while chrome alum is deep purple in colour and ferric alum in pale violet and has application in dyeing of fabrics, tanning of leather, water clarification, as an astringent or styptic, in medicinal and non medicinal applications.

In this experiment you will transform aluminum metal into alum (aluminum potassium sulphate dodecahydrate), $KAl(SO_4)_2 \bullet 12H_2O$ taking advantage of aluminum's chemical properties. Alum would not typically be synthesized from pure metallic aluminum owing to the high cost and significant environmental issues associated with the production of the metal but this synthesis provides an ideal showcase for aluminum chemistry and reactivity. Aluminum cans are made from fairly pure aluminum but for decorative paint coatings on the external surface and a thin coating of plastic on the inside to prevent dissolution of the aluminum can in acidic beverages. You must first mechanically remove these coatings prior to completely the chemical transformation of aluminum to alum.

Aluminum metal has a high negative standard reduction potential, thus any exposed surface of aluminum metal will be readily oxidized by $O_2(g)$ to form aluminum oxide, $Al_2O_3(s)$. A thin layer 10^{-4} to 10^{-6} mm impermeable layer of oxide will form which forms a protective coating on the aluminum metal surface. The first step in the process is dissolution of the oxide layer and the subsequent oxidation of aluminum metal, in a basic environment (KOH), to form the soluble aluminate anion. Water is reduced.

Addition of a slight excess of sulphuric acid, H_2SO_4, to the products neutralizes the excess KOH and also the for OH^- groups on the tetrahydroaluminate complex ion to form soluble aluminum sulphate. The resulting solution contains K^+, Al^{3+}, and SO_4^{2-} ions in the proportions in which they are found in alum: $KAl(SO_4)_2$, and on sufficient cooling, crystals of the hydrated potassium aluminum sulphate, or alum, will form.



Experimental procedure

Prelab exercise: Preparation of aluminum sample prior to attending the laboratory

You will need a pop can to be used as the source of aluminum metal for this experiment.

Cut a piece of aluminum 10 x 10 cm from an aluminum pop can –once cut it will be sharp so be careful! Scour both surfaces of the aluminum square until the aluminum is clean and bright. Rinse and wipe the aluminum. Cut the 10 x 10 cm square into smaller squares each about 0.5 cm x 0.5 cm. Bring your sample to the laboratory.

In laboratory procedure:

You are to prepare two (2) samples.

Both Samples: In a 150 ml beaker, add between 0.5 and 0.6 g of aluminum and record mass to nearest milligram. Carefully add 25 ml of 1.4 M KOH to the beaker containing aluminum. Heat at low heat, stirring occasionally, but do not allow the sample to boil.

Sample 1:
Once aluminum has dissolved, remove beaker from hot plate. Add 10 ml of 9 M H_2SO_4 carefully but rapidly to the beaker, stirring steadily. Stir the solution thoroughly. If white crystals are visible, return beaker to hot plate and stir until dissolved. Allow beaker to cool so that it may be safely handled, but is still warm to touch and all white crystals dissolved.

Set up a glass funnel with appropriate filter paper and place stem in a 150-ml beaker. Secure filter paper with a small amount of water and discard the water that runs through the filter to the beaker. Filter your sample while still warm. Allow filtrate to cool. Use a 600 ml beaker and prepare an ice bath. Place the smaller 150 ml beaker in the ice bath, stirring frequently.

Sample 2:
Once aluminum has dissolved, remove beaker from hot plate. Set up a glass funnel with appropriate filter paper and place stem in a 150-ml beaker. Secure filter paper with a small amount of water and discard the water that runs through the filter to the beaker. Filter your sample while still warm.
Add 10 ml of 9 M H_2SO_4 carefully but rapidly to the warm filtrate, stirring steadily. Stir the solution thoroughly. If white crystals are visible, return beaker to hot plate and stir until dissolved.

Allow filtrate to cool. Use a 600 ml beaker and prepare an ice bath. Place the smaller 150 ml beaker in the ice bath, stirring frequently.

Both Samples:
Set up a vacuum filtration apparatus with the Buchner funnel an appropriate suction flask and appropriate filter paper. Moisten filter paper to hold it in place. When crystallization is complete, measure the solution temperature (less than 6 ^oC). Transfer all crystals to the Buchner funnel.

Add two portions of methanol in succession to wash the excess H_2SO_4 out of the crystals. Continue to draw air through your sample for 10 to 15 minutes.

You will now determine, by difference, the mass of your crystals and % yield. To a previously weighed clean dry 150 ml beaker transfer the crystals and filter paper. Remove crystals from filter paper and reweigh the beaker.

Report

Calculate the percent yield for each sample.

Your discussion should include chemical reactions for each step of the synthesis as well as an explanation of the sample yield for each method. In addition, explain how you would confirm that the recovered solid is alum.

Questions

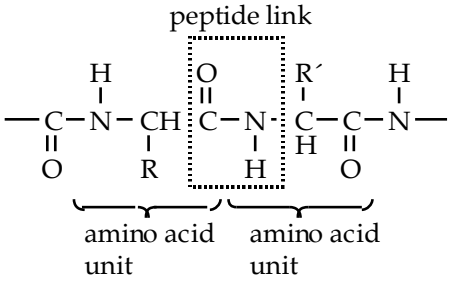
1. If acid is added slowly, a white precipitate appears and subsequently disappears. What is the white precipitate? Support you answer with appropriate chemical equations.
2. Experiments show that the solubility of alum in 25 ml of 1.4 M KOH and 10 ml of 9 M H_2SO_4 is approximately 1.0 g at 1.0 ^oC and 1.7 g at 6.0 ^oC. Estimate the amount of alum left in the 65 ml of a solution chilled to 4 ^oC. What might you do reduce the amount of alum remaining in solution?
3. Provide examples of an i) acidic oxide; ii) basic oxide; iii) amphoteric oxide (other than Al_2O_3) and iv) neutral oxide.

Experiment V

Analysis of Organic Nitrogen in Water by Kjeldahl Distillation

Ammoniacal and organic nitrogen

In environmental chemistry, organic nitrogen is nitrogen in the -3 oxidation state which is part of an organic molecule. An example is the amino acid alanine, $H_2NCH(CH_3)COOH$. Proteins are constructed by the formation of peptide links between the amino groups and the carboxylate groups of this and other amino acids:



The most abundant source of nitrogen for protein synthesis is atmospheric N_2. However, the N_2 molecule is very stable and unreactive, such that most organisms cannot use it in this form. Nitrogen is only directly assimilated by plants in the form of nitrate, NO_3^-. Animals are not capable of using either N_2 or nitrate directly as the source of nitrogen for protein synthesis. With the exception of ruminants, animals depend on plants, or other animals that feed on plants, as their source of protein.

While nitrate salts are commonly used as fertilizers, and their possible contamination of drinking water is a health concern (see Experiment II), ammonia and ammonium salts can also be applied to soils to supply plants with nitrogen for protein synthesis. The ammonia is oxidized by soil bacteria to nitrate. Urea is often used as a fertilizer because ammonium is released gradually as the urea hydrolyzes, eq 5.1.

$$H_2NC(O)NH_2 + 3\,H_2O \leftrightarrows 2\,NH_4^+ + HCO_3^- + OH^- \qquad (5.1)$$

Application of excess fertilizer leads to the contamination of water by ammonia. Ammonia also occurs naturally in surface and wastewater. Deamination of proteins leads to the formation of urea, which is excreted in the urine and then hydrolyzed to ammonia.

In aqueous solution, ammonia exists in equilibrium with its protonated form, eq 5.2.

$$NH_3 + H_2O \leftrightarrows NH_4^+ + OH^- \qquad pK_b = 4.74 \qquad (5.2)$$

Ammonia concentrations encountered in water may vary from less than 10 □g ammonia-N/L in natural surface and groundwaters to more than 30 mg/L in wastewater. Neutral ammonia is toxic to fish above 0.2 mg/L (although the ammonium ion is not). Therefore ammonia toxicity is not a problem in natural waters with pH below 8 and ammonia-N concentrations

less than 1 mg/L. However, ammonium is oxidized by autotrophic bacteria to nitrite and nitrate in a process which consumes oxygen, eq 5.3 and 5.4, so the discharge of ammonia/ammonium has the potential to seriously deplete dissolved oxygen in natural bodies of water.

It is also important to control the ammonia concentration in water treatment plants. In the disinfection of drinking water, chlorine reacts with ammonia to form combined chlorine. In Ottawa-Carleton water treatment facilities, ammonia is intentionally added to the chlorinated water to form nontoxic monochloramine, which is a longer-lived disinfecting agent than aqueous chlorine itself. The low concentration of chloramine in our drinking water thereby protects us from water-borne pathogens. Virtually no free chlorine residual is obtained until the ammonia has been completely oxidized. The amount of ammonia present in the water determines the amount of chlorine required to obtain a free (not combined) chlorine residual. The addition of enough chlorine to oxidize all the ammonia is called *breakpoint chlorination* and is performed in water disinfection to destroy bacteria.

Ammonia can be analyzed directly using an ammonia-selective electrode. More commonly, organic nitrogen and ammonia are analyzed together by the Kjeldahl (pronounced Shel´-dal) distillation method. The total is referred to as the *Kjeldahl nitrogen*. We will analyze the Kjeldahl nitrogen present in a synthetic urea solution.

Kjeldahl method

In the Kjeldahl method, the organic components of an aqueous solution are *digested*, or completely oxidized, to CO_2 and H_2O. Any nitrogen contained in the organic compounds is released as ammonia, which is immediately protonated in the strongly acidic digestion medium. Then the digested solution is made alkaline, causing the nonvolatile ammonium ion to be converted into volatile ammonia. The NH_3 is distilled out of the solution and captured in a boric acid solution, eq 5.3. The amount of ammonia in the distillate is determined by back titration of the borate anion with a strong acid, eq 5.4.

$$NH_3 + H_3BO_3 \leftrightarrows NH_4^+ + H_2BO_3^- \quad \text{capture of ammonia} \qquad (5.3)$$

$$H_2BO_3^- + HCl \leftrightarrows H_3BO_3 + Cl^- \quad \text{back-titration} \qquad (5.4)$$

The Kjeldahl method uses sulfuric acid as the oxidizing agent. During the heating period, excess water is first expelled, leaving concentrated sulfuric acid to attack the organic matter. When the sulfuric acid starts to boil, copious white fumes of SO_3 form in the flask. As the digestion proceeds, bubbles of CO_2 and SO_2 form due to oxidation of organic matter and reduction of H_2SO_4.

Experimental procedure

THIS IS A LONG EXPERIMENT – YOU MUST BE PREPARED AND MAKE GOOD USE OF YOUR TIME.

Caution: Concentrated sulfuric acid is highly corrosive. Avoid contact with the skin. Fumes

of SO_3, SO_2 and NH_3 are irritating. Do not breathe them. Perform this experiment in the fumehood and wear your safety glasses at all times.

This experiment will be performed by pairs of students. During the waiting period of the digestion step, perform the indicator tests and set up the distillation-titration apparatus described in Step 2. Ask your demonstrator to check your setup before beginning the distillation. Check for bubbles in the burette stopcock before starting the titration.

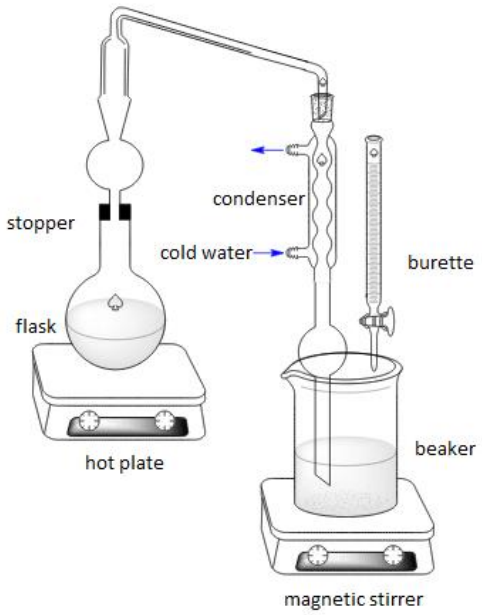
Step 1. Digestion

In each of two 50 mL beakers, place 5 mL of conc. H_2SO_4. Add 2.0 mL of the urea sample solution by pipette. In a fumehood, heat the solutions to boiling using a hot plate. Boil for a total of 30 minutes. DO NOT ALLOW SOLUTIONS TO BOIL DRY. Allow the solutions to cool.

Step 2. Simultaneous distillation-titration

Test the indicator solution: put approximately 1 mL of 0.1 M HCl in a test tube and add a few drops of the Tashiro indicator (a mixture of methyl red and methylene blue). Repeat in a second test tube with 0.1 M NaOH. Now put 1 mL of the boric acid solution in a third test tube, add a few drops of indicator and NaOH dropwise until you find the colour of the endpoint. Record the colours which correspond to acidic, neutral and basic conditions.

Set up the distillation apparatus as shown in the diagram and have your demonstrator check it before attempting the distillation. The tip of the condenser should be completely immersed in 100 mL of boric acid. Add 10 drops of Tashiro indicator to the boric acid. Adjust the colour of the indicator by adding NaOH dropwise until the solution is green, then adding HCl dropwise until you reach the greyish endpoint colour. (If the colour change is not clear, add more indicator.) Fill the burette with standardized 1 M HCl and record the initial volume.



Add 50 mL of distilled water to the Florence flask then *slowly* pour the digested solution into the water. Rinse the digestion flask with water and pour the rinse water into the Florence flask. Make up the volume to approximately 250 mL with distilled water. Add a few drops of phenolphthalein and a few boiling stones. Add 50 mL of conc. NaOH and install the stopper immediately so that the volatile ammonia does not escape. If the solution is basic enough, it will be pink (at least transiently). Heat the solution vigorously with the Bunsen burner. After several minutes, bubbles will appear in the boric acid solution at the base of the condenser. When the colour of the boric acid solution changes, add HCl dropwise from the burette to bring the solution back to its original colour. (The solution may backflow into the condenser. Keep an extra beaker of boric acid on hand and add as needed to keep the tip of the condenser immersed in boric acid at all times to prevent ammonia from escaping.)

Continue the addition of HCl throughout the distillation. Liquid water may form in the tubing; this will not affect your result. The distillation is finished when the endpoint colour persists for at least 5 minutes. Remove the condenser from the boric acid solution and turn off the Bunsen burner. Record the final volume of HCl in the burette.

Repeat the distillation on the second digested sample and average the results.

Report

Calculate the amount of HCl consumed in the titration and the quantity of NH_3 displaced into the boric acid solution. Determine the urea concentration in your unknown sample.

Discussion

Report the concentration of urea in the sample solution. What is a practical application of the Kjeldahl method? What are the limitations of the method for this application? How could it be improved and/or suggest an alternate method ?

Questions

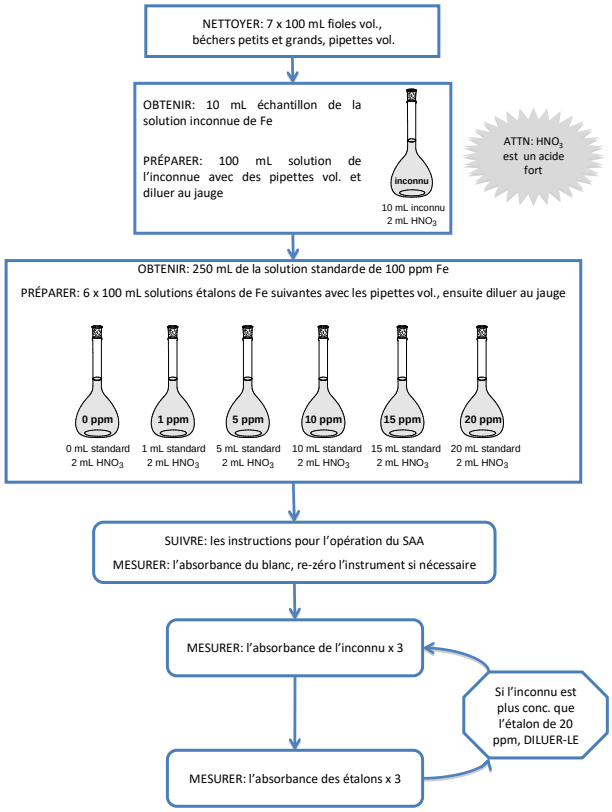
1. What is the purpose of the "digestion" and how is it accomplished?
2. An alternate protocol calls for the distillation of the ammonia into an excess of a standard solution of H_2SO_4 followed by titration with standard NaOH to determine the concentration of H_2SO_4 remaining. (i.e. back titration) What advantages does the simultaneous distillation/titration with boric acid offer?
3. Can this method be used to analyze organic nitrogen in the form of pyridine?

Check-out

Before leaving the lab, verify that your locker contains all the equipment listed at end of manual. Each item should be clean and free of chips or cracks. Replace any items which are broken. Show the locker checkout sheet to your demonstrator before leaving.

Appendix A1

Example of Pre-lab flow chart



Hazardous materials:

HNO_3: a colourless to yellow strongly oxidizing liquid. Conc. Nitric acid (liquid and vapour) is corrosive to eyes, skin and mucous membranes. Contact can burn. Inhalation can cause coughing/shortness of breath. Safety: goggles/face shield; lab coat; fume hood for concentrated acid. Spill: neutralize with sodium bicarbonate or other suitable material. Skin contact: wash for 15 minutes; eyes: wash eyes for 15 minutes with eyelids open, seek medical attention.

Waste: Disposal of waste solutions dependent on nature of cations and anions – all must be deemed no hazardous if solution is to disposed of in sink.

Appendix A2
UV-visible spectrophotometer

Spectrophotometry refers to the use of light to measure chemical concentrations, and a spectrophotometer is an instrument containing a light source and a detector.

In a UV-visible spectrophotometer, light from a continuous source such as a tungsten lamp (in the visible) or a deuterium lamp (in the UV) passes through a monochromator, which selects a narrow band of wavelengths from the incident beam. This *monochromatic* light passes through the sample and emerges to be detected at a photomultiplier tube.

The light source is an ordinary tungsten lightbulb, which emits light across the entire visible spectrum and some of the UV. The light is dispersed into its component wavelengths by a diffraction grating, so that only one small band of wavelengths passes through the sample. The detector is a phototube which creates an electric current proportional to the radiant power of the light striking the tube. The output is expressed on a meter which reads in both transmittance and absorbance. It is important to read from the correct scale.

The relationship between concentration and the amount of light absorbed is given by Beer's law:

$$A = \log I_0/I = \varepsilon bC \qquad \text{(A2-1)}$$

where I_0 is the intensity of light entering the solution, I is the intensity of light exiting the solution, ε is a constant called the molar extinction coefficient, which depends on the molecule and the wavelength, b is the pathlength of the light passing through the solution and C is the molar concentration of the absorbing species in solution. The quantity $\log I_0/I$ is called the *absorbance*, A. The greater the absorbance, the darker the solution appears in colour. For a given species, at fixed wavelength and pathlength, the absorbance is directly proportional to the concentration. The transmittance T is defined at I/I_0, so that
$A = - \log T$.

In practice, I_0 is measured as the light intensity passing through a reference solution. This procedure compensates for reflection, scattering and absorption of light by the reference solution and the sample holder.

Please circle course number. (2353, 2753, 2354, 2754)
Student (1)_____
Student (2)_____

Locker # _____
Demonstrator _____

Drawer	IN	OUT		IN	OUT		IN	OUT
Wire gauge	2		Filter funnel	2		Volumetric pipet 1 ml	2	
			Powder funnel	2		Volumetric pipet 5 ml	1	
Clamp holder	2		Sintered glass funnel	2		Graduated pipet 5 ml	1	
Support ring	2		Large watch glass	4		Glass rod with policeman	2	
Rubber tubing	2		Small wash glass	6		Spatula/Scoopula	2 ea	
Bunsen burner	2		Wash bottle	2				
Crucible tongs	2		Pipet bulb	2		Suction adapter	1	
Buchner funnel	1							

BOTTOM SHELF	IN	OUT	TOP SHELF	IN	OUT
			Universal clamp	2	
Suction flask 500 ML.	2		Graduated Cylinder 10 ML	2	
Erlenmeyer flask 50 ML.	2		Graduated Cylinder 50 ML	2	
Erlenmeyer flask 250 ML	8		Graduated Cylinder ML	2	
			Beaker 50 ML	4	
Volumetric pipet 10 ML	2		Beaker 150 ML	2	
			Beaker 250 ML	4	
Volumetric flask 100 ML	20		Beaker 400 ML	4	
Volumetric flask 250 ML	4		Beaker 600 ML	2	
Volumetric flask 500 ML	2		Beaker 1000 ML	2	
Bottle 1000 ML (brown)	4		Rack for 50 ml plastic centrifuge tubes	2	
Bottle 1000 ML NALGEN	4		50 ml plastic centrifuge tubes	20	

Note the two 50 ml burets and one 20 or 25 ml volumetric pipet in the door - BE CAREFUL when closing the door.
You must show your locker to the demonstrator before leaving.