

# CHM 317H1 S

# Instrumental Analysis

# Laboratory Manual



**Department of Chemistry**  
**University of Toronto**

**2017 – 2018**

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**Web resources:**

1. Statistics and Regression Tutorial  
<http://www.chem.utoronto.ca/coursenotes/analsci/stats/>
2. Pipette Technique  
<http://www.chem.utoronto.ca/coursenotes/analsci/mm/index.html>
3. Instrument “Virtual Tours”  
<http://www.chem.utoronto.ca/coursenotes/analsci/instruments.html>

## INTRODUCTION

This third-year laboratory course in Analytical Chemistry builds on the skills and techniques encountered in the second-year course. It consists of five groups of experiments, each featuring different instrumental techniques. Two will extend content covered previously in the second-year course (infrared and flame atomic spectroscopy), one will introduce fluorescence spectroscopy, and two will be devoted to gas and liquid chromatography. Evaluation will be based on laboratory performance and four formal reports, one for each group of chromatography experiments, and two out of the three *groups* of spectroscopy experiments. You will work in groups of two or three each week, but are responsible for writing and submitting your *own* report based on the technique used and the data obtained jointly by you and your partner.

You will be required to maintain a hard-bound laboratory notebook according to the principles of good laboratory practice (GLP). It is also important that you pay attention to significant figures, and perform appropriate error analysis for all experiments; do this in your lab notebook as you perform the experiments.

- **All students** are expected to be familiar with, and abide by, all applicable health and safety regulations. This includes the wearing of safety goggles and lab coats, and the safe handling and disposal of all chemical substances. Relevant details are included in sections 6 and 7 of the introduction to this lab manual.

You will be expected to prepare yourself in advance for each lab session by familiarising yourself with the techniques and instrumentation (from the course text and other resources as necessary), and obtaining relevant safety and material properties information for the substances being handled in the lab. This should be recorded in your lab book ahead of time, and checked by your demonstrator at the start of each group of experiments.

Students should attend every laboratory session, and are responsible for cleaning up all glassware, chemicals, and materials used in the course of their experiments. Punctuality is essential; students who are unavoidably detained should make every effort to let the lab staff (and their partners!) know when they expect to arrive. Attendance records will be kept; students who are obliged to miss a lab due to illness or family emergency should contact the lab instructor with appropriate documentation as soon as possible. Further information, including supplementary material for this manual, can be found at:

<http://www.chem.utoronto.ca/coursenotes/CHM317/>

**1. Staff**

Lab Instructor:	Dr. David Stone	LM 218	416-946-0293
Lab Supervisor:	Dr. Jared Mudrik	LM 9	416-978-3599
Lab Office (during lab Mon–Weds)		LM 6	416-978-5060

To contact the lab instructor or your TA, please use the course communication tools on the portal. Students are advised to discuss each experiment – especially any questions they might have regarding calculations and report requirements – with their TA *during the laboratory sessions and before the report is due*.

**2. Laboratory Schedule**

The schedule of experiments is available on the public course web site (<http://www.chem.utoronto.ca/coursenotes/CHM317/timetable.html>); you should check this page and the lab manual page regularly for updates and additional information. Please note that some experiments require you to plan what you will be doing *ahead of time*. This includes preparing and bringing in your *own* samples: always read ahead so that you have adequate time to do this!

**3. ATTENDANCE AND MISSED LABS**

You are expected to attend every lab, and be *ready to start at 9:00 am sharp*. Lateness penalties apply in increments of 5 minutes from 9:10 to 9:30; this is because your lateness will delay other members of your group, who will be depending on your cooperation. Consideration will be granted for circumstances beyond your control, however you are strongly encouraged to plan to arrive *early*. Students who arrive more than 30 minutes late *without* good reason will lose their performance marks for that particular experiment.

**If you are unable to attend due to illness:** Please make every effort to notify the lab staff as well as your partner or team members. This is important, as your fellow students need to know if you will be there. You will need to have a medical professional complete a “Verification of student illness” form: please see <http://www.illnessverification.utoronto.ca/> for details.

Students who miss a lab *may* apply to the course instructor for permission to perform that experiment with a different group in an alternate lab section (*i.e.* on a different day of the week.) All such applications must be made *as soon as possible*, and will be accommodated only if it is possible. Please note that you *must* inform the lab coordinator and your lab partners of your absence *even if* you declare a student absence on ROSI, since the system does *not* relay such notifications to your instructors.

**4. Expectations and Grading Policy**

You are expected to come to each lab fully prepared for that day’s experiment, with all your calculations from the previous experiment completed and written up *in your lab notebook*. You will be expected to demonstrate that you know what chemicals you will be working with, what hazards may be associated with those substances in the forms and concentrations you will be using, and the relevant safety procedures to be followed during your experiment. You are expected to:

- Arrive *on time* and fully prepared (includes eating a good breakfast!)
- Work cleanly, safely, and efficiently, and communicate clearly
- Co-operate fully with group members, demonstrators, and staff
- Share work equally with your partners, taking turns to operate instruments
- Clean up completely and properly – this is a safety issue!
- Leave *on time* – the lab is used by other courses and research students

**The more time you spend in preparation, the less time it will take to obtain good results and complete the experiment!**

The total mark for the laboratory portion of this is broken down into a weekly preparation/performance component, weekly results summaries, and four formal reports, as summarised below. Your weekly performance mark will be determined, in part, by how well you meet expectations. More specific information is provided in the following sections. Your total lab mark will count for **40%** of your final course grade.

*Laboratory Course Marking Scheme*

Item	Marks
Writing (WIT) grade for first week's experiment	20
Four cumulative formal reports (55 marks each): FTIR Spectroscopy Fluorescence Spectroscopy Gas chromatography Liquid chromatography	220
Weekly performance mark (12 marks $\times$ 10)	120
Weekly executive summary (4 marks $\times$ 10)	40
<b>Total Laboratory Mark</b>	<b>400</b>

#### 4.1 Weekly Performance Marks

You will also be required to maintain a full record of your activities, results, and calculations in your lab notebook. This *must* be kept up-to-date at all times, and will be inspected by your demonstrator. In keeping with the principles of Good Laboratory Practice (GLP), you must have your notebook validated by your demonstrator before you leave each week. Part of your weekly mark is based on your notebook; detailed instructions follow. Note that performance marks may be voided if a required lab report is not submitted for grading.

Preparation (max. 4 marks/week):

You are expected to prepare yourself for each experiment by reading through the instructions, determining the chemical & physical properties of the substances you will be using, identifying any particular hazards, and calculating the weights and dilution factors

needed to make the solutions required. Your notebook should contain the following when you arrive in the lab to perform your experiment:

- The title of the experiment and brief description of aims/objectives
- A table of relevant chemical & physical data, and any specific hazards
- Any necessary calculations (include relevant mathematical equations)
- A summary of the procedure to be followed – see the example in sect. 8

**Note:** hazard tables printed from a web site and simply pasted into your notebook are *not* acceptable! Summarize the information including *only* the details relevant to the *physical forms and concentrations* you will actually be using in the lab.

Performance (max. 4 marks/week):

During the lab, you will be assessed on how well you conduct the experiment. This includes various aspects, such as proper use of volumetric glassware, recording of data in your lab notebook, efficient use of chemicals and time, safe working practices, following lab procedures, *etc.* You are required to present your lab notebook to your demonstrator at the end of every period to be validated. In particular, the demonstrators will be checking that you have recorded what you actually did during the lab, as well as calculating weights and actual solution concentrations, and recording all relevant data. Please see section 8 ('Your Laboratory Notebook') for further details as to what is expected of you.

Calculations (max. 2 marks/week, excluding first and last weeks of the lab):

You should always plot calibration data as you acquire it, and check that your sample measurements yield reasonable results as you go. All follow-up calculations *must* be completed *in your lab notebook* during the week following the experiment, so that you have a permanent record of what you have done. This includes inserting copies of any calibration curves, regression data, and analyte concentrations determined for the preceding week's experiment. Your demonstrator will check that you have done this during the following week's lab. An independent researcher reviewing your lab notebook should be able to determine exactly what you have done simply by reading your account and calculations.

Co-operation (max. 2 marks/week):

Since you will be working with a partner throughout this course, it is important that you work together well, making the most efficient use of your time. For example, if a number of different solutions have to be prepared, one of you can be running or preparing the instrument while the other makes solutions; you should switch duties periodically so that you *both* have a chance to run the instrument. You are also expected to co-operate with the lab staff and other students in the course, since you will often be sharing balances, bench space, *etc.* with them.

Punctuality and Clean-Up:

Because there is limited space, and other courses and researchers also use the facilities, it is essential that you both arrive on time and clean up before leaving. You should arrive by 9:00 am in order to be ready to start promptly at 9:10! **Marks may be deducted** for lateness (up to a maximum of 5) and failure to clean up properly (up to a maximum of 5).

All equipment used must be cleaned, and all chemicals returned to the appropriate locations, by the end of the laboratory period.

If you are unavoidably detained (*e.g.* by the vagaries of public transit in winter), please try to let the lab staff and your partner know as soon as possible; if necessary, you can call the lab and leave a message.

- ☛ **Students who are habitually late** will, at the discretion of the lab instructor, receive a performance mark of zero for each lab (including the report) for which they arrive late. They may also be refused permission to perform the lab, and will *not* be given the opportunity to perform a make-up lab.

## 4.2 Writing Initiative (WIT) Component

The formal reports for this course incorporate multiple experiments, as would be the case for an extensive method development or review journal article. As such, they can be quite daunting to write, especially for students without much practice in earlier courses. This course is part of the Writing Instruction for and by TAs (WIT) program run within the Faculty of Arts & Science. The aim of this program is to help students develop better writing skills within the context of the subject they are studying – in this case, chemistry. The WIT component of your lab grade is a mandatory, graded writing exercise that will help you develop your writing skills as well as get you started on your first formal report. Although the WIT component only represents a small portion of your final lab grade, students who do *not* participate will *not* be allowed to re-submit a failing report as set out below.

### WIT Component for First Experiment (20 marks):

To help you prepare your first formal report, you are **required** to write up a report for your *first* experiment, and submit this to your TA at the start of the next lab period. You will receive this back the following week with comments on your writing that you should use when writing your final report.

**What to include:** Your draft report should include all the elements required for a full report, but *only* for the content relevant to your *first* experiment. This includes: a title page with signed academic integrity pledge, an abstract, a short introduction, experimental details, your results, a complete discussion, a conclusion, and any relevant references. See section 5, “Formal Reports” for more detailed information on your report. For the purposes of *this* exercise *only*, your abstract should be ~50-100 words, with an introduction of up to 300 words, discussion of up to 500 words, and conclusion of up to 150 words maximum.

**How your writing will be evaluated:** The WIT component mark will be awarded based on the completeness of your submission and the degree to which it meets the requirements set out in section 5, *not* on the actual results of your experiment. You will receive feedback from the TA *on your writing* the following week, in order to help you better prepare your first full report. To earn full marks on subsequent reports you must implement your TA’s feedback from your draft when writing your formal reports.

### 4.3 Formal Reports (Four worth 55 marks each):

**Due date and late penalty:** Formal lab reports are generally due at the *start* of the lab period following completion of each group of experiments, as indicated on the lab timetable. Please check with your lab TA for specifics. If you believe you will not be able to complete your report on time, you *must* contact the lab instructor at least two days before the due date if you wish to receive consideration for late submission. In the event of a medical or family emergency, you will need to provide the same documentation as required for a missed lab. A late penalty of 5% of the maximum possible mark per business day past the deadline applies *unless* you have such documentation or arranged an extension with the lab instructor. Reports will generally be marked and returned (with comments) within two weeks of submission.

**When to write your reports:** Although you will not submit your actual formal reports until you have completed all the experiments in each section, you should do all calculations and write up the relevant discussion *each week*. This will give you time to clarify any issues with your lab partners, and seek advice from your TA should you have difficulty with the material. To help you stay on track, this will form the basis of the weekly *executive summary* you will need to submit.

**Marking:** Students who earn a mark of less than 50% on their *first report* may request permission **from the lab instructor** to re-write and re-submit it *only* if they participated in the WIT component (see above). Note that a limit of 80% of the maximum possible grade applies on any resubmitted reports. Students wishing to re-submit their first report should contact the lab instructor no later than 1 week after receiving their graded report.

Further details concerning the format, content and marking of reports are provided in section 5. Please talk to your demonstrator or lab instructor *before* the deadline if you have any problems or questions regarding your data, calculations, theory, discussion, *etc.*

- ☞ Do not wait until you have completed all the experiments to start writing your report, or you will not have enough time to complete it.

### 4.4 Departmental Policy on Plagiarism:

Students *must* read the departmental policy on plagiarism, as well as the examples provided *via* the departmental web site (see following page and link below). All students will be required to sign a statement to the effect that they have read and agree to abide by this policy. You are reminded that other student's lab reports are considered to be *unauthorized study aids* under the terms of University and departmental regulations and policies, and should not be used when preparing your own reports.

<http://www.chem.utoronto.ca/undergrad/plagiarism.php>

With respect to this course and the following departmental policy, it is expected that students will share data and discuss experimental procedures, as well as any problems encountered, with their partners. When it comes to performing any calculations and writing up your results and discussion, however, all work should be your own.

**DEPARTMENT OF CHEMISTRY, UNIVERSITY OF TORONTO**

**POLICY REGARDING LABORATORY ACADEMIC DISCIPLINE**

The following regulations are quoted from Section B of the University of Toronto Code of Behaviour on Academic Matters:

B.i. 1. It shall be an offence for a student knowingly:

- (b) to use or possess an unauthorized aid or aids or obtain unauthorized assistance in any academic examination or term test or in connection with any other form of academic work.
- (d) to represent as one's own any idea or expression of an idea or work of another in any academic examination or term test or in connection with any other form of academic work, *i.e.* to commit plagiarism.

Wherever in this Code an offence is described as depending on "knowing", the offence shall likewise be deemed to have been committed if the person ought reasonably to have known.

In response to these regulations, the Faculty of Arts and Science enforce the following policy:

**Student laboratory reports (both past and present) are unauthorized aids, and making use of them *in any way* constitutes an academic offence**

Although undergraduate students may discuss experimental data with one another, an important consequence of the policy stated above is that every student is required to write an individual report. **CONSULTATION WITH OTHER STUDENTS OR COLLABORATION AMONG STUDENTS IN THE WRITING OF LABORATORY REPORTS IS NOT PERMITTED** except as officially sanctioned and **set out in writing by the course co-ordinator or laboratory instructor**. This policy also applies to any required pre-laboratory preparation.

**An academic offence set out in the Code of Behaviour pertaining to plagiarism *in any report* will lead to a mark of zero for the entire laboratory component of the course**

For more information (including examples of plagiarism and additional Faculty sanctions for academic offences), please visit our web site:

**<http://www.chem.utoronto.ca/undergrad/plagiarism.php>**

**Please refer any questions concerning this policy to the laboratory instructor.**

## 5. Formal Reports

During the course of this lab, you will write a cumulative formal report for each chromatography technique, and cumulative formal reports for two of the three spectroscopic techniques, for a total of four formal reports. These reports give you an opportunity to relate theory to practice so that you gain a greater understanding of how the technique works, and what its particular advantages and areas of application are. The style of report is that for a full analytical research article or technical review, giving you additional practice in scientific writing. Since the reports count for the greatest portion of your laboratory mark, it is important to plan ahead so that you are not trying to do everything at the last minute. This is why, for example, you are required to do all the calculations and graphs for each experiment *before* the next lab. The following sections provide more detailed explanations of what is required for a successful formal lab report.

### 5.1 Writing Support Services

If you have not had much writing experience, or have found writing assignments in earlier courses difficult, you are *most strongly advised* to look into the services offered by the University's Writing Centres *especially* for your first report. In addition to numerous helpful on-line resources, staff at the centre can guide you through the research, writing, and editing stages of your report. The main writing support site for the university can be found at:

<http://www.writing.utoronto.ca/>

This includes a listing of the various writing centres, where you can receive individual assistance with writing, as well as numerous on-line resources and advice. Students who have not had much experience in writing formal science reports and papers are strongly advised to contact one of the writing centres as early as possible during the semester.

### 5.2 Format and General Structure

Each formal report should be printed using a *clearly legible* 12-point font: good examples include Garamond, Times, and Arial. *Do not use fancy calligraphic or handwriting fonts!* **Format your report as single column**, in double- or 1.5-line spacing (to allow room for your TA to write comments). Check with your TA as to whether they have a preference for single- or double-sided reports. You should write in the traditional scientific style (third person passive voice, *e.g.* "The solutions was..." and **not** "I did this to the solution") appropriate for a full research or review paper. (This would be ~5-6 printed pages in a journal such as the *Analyst* or *Analytical Chemistry*). Your report should consist of the following main sections, which are discussed in more detail below:

- Title page, including:
  - Name, demo group, and date
  - Academic honesty pledge
  - Your signature
- Abstract (75–125 words, max. 150)
- Introduction (max. 500 words on theory, 500 words on instrumentation)
- Experimental

- Results & Discussion
- Conclusions (100–200 words, max. 250)
- References
- Appendices

### Grammar and Mechanics: How to ensure your report is well-written

- Proof-read your report before submission. Pay particular attention to sentence structure; an independent reader should be able to follow the sequence and logic of your thoughts, observations, and conclusions
- Use the spelling and grammar check tools that come with your word processing software: they will catch *many* common errors, although they won't detect errors in logic!
- Read what you have written out loud; you should be able to do so without stumbling over awkward phrasing, or pausing while you try and decide what you actually meant.
- Remember that your TA can *only* mark what they read, not what you meant to write!

### Word Count and Format

- For *each* section, please include your actual word count at the end of the section heading. A deduction of up to 10% of your mark will be made for work exceeding the stipulated length. Number all figures, equations, and tables, and make sure you provide a clear yet concise caption for each one.
- All relevant sources and quotations used in the report *must* be referenced; use sequentially numbered end-notes for your references, and follow a consistent format for all citations.
- Use the American Chemical Society reference format. Remember that the primary aim of any reference format is to make it easy for others to find the work you are citing: this means that author, name of publication, year of publication, and page numbers must be clear and correct.

ACS Style Guide Chapter on Citations:

<http://pubs.acs.org/userimages/ContentEditor/1246030496632/chapter14.pdf>

#### 5.2.1 Title Page

- Clearly state the title of the formal report, as well as your name, student number, demo group number, and due date for the report.
- You **must** (according to departmental policy) include the following statement on your title page, which you should also *sign and date in ink*:

“I certify that this submitted laboratory report represents entirely my own efforts. I have read and understand the University of Toronto policies regarding, and sanctions for, plagiarism”

Your title should reflect the overall aim of the experiment(s), referencing the type of sample or instrumentation involved; do not simply write “Experiment A” or “HPLC Report”. For example, if you had performed a set of experiments involving the analysis of metal ions in solution using molecular absorption spectroscopy, your title might be:

“Factors affecting the simultaneous determination of cobalt(II) and chromium(III) in aqueous solution using UV-visible spectrophotometry”

### **5.2.2 Abstract (2 marks; 150 word maximum)**

An abstract (or in business terms, an executive summary) should communicate simply and clearly what the body of the report is about, including the nature of the work undertaken and the key findings from the results. For journal articles, the abstract is an open advertisement for the paper, and may well be the only part of the article that gets read by a wide audience. A good abstract therefore needs to be informative, engaging, and contain the key words that would be used by someone searching for information on the topic. It should describe the aim of the work, list the techniques used, and summarize your conclusions. It is often easier to write the abstract once you have finished the rest of the report.

For these reports, your abstract should be ~ 75–125 words (150 maximum) in length. If your experiment included quantitative analysis, report the type of sample, analyte, and concentration (properly rounded) with uncertainty and correct units. Identify any important trends observed in your experiments, such as the effect of temperature or interfering species present in your sample; identify how you will explain the results within the report. If your experiment involved a comparison of techniques or conditions, briefly list the basis for the comparison. Do *not* include specific procedural details, such as the make and model of any instrumentation used or the concentrations of calibration standards.

### **5.2.3 Introduction (8 marks; max. 500 words theory & 500 words instrumentation)**

An introduction explains the aim of your work, the reasons for conducting it, and provides the background (both theory and practice) required to understand the experiments, interpret the results, and follow the discussion. In research articles, this would include a brief summary of prior related work; review articles would usually reference seminal papers and earlier reviews, along with their scope.

Where a new technique or theory is described, a full explanation and derivation of any equations would be required; for established techniques and theories, only a summary of the key points and equations of relevance is needed. Where a new instrument is described, or where the operation of the various internal components is essential to understanding the experimental results, clear diagrams are required; similarly, the relationship between the instrument signal and the sample concentration should be established. Make sure you include citations for key concepts, equations, and instrumentation (but see the notes on citation below, section 5.3). Avoid quoting extensively from source material: put as much as possible *in your own words*.

For these reports, your theory section (max. 500 words) should include any relevant equations to help you describe the physical operation of the instrument. Your description of the instrumentation (max. 500 words) should include a block diagram of the *generic* instrument; other diagrams detailing relevant components of the instrument are highly encouraged. Drawing them yourself can help familiarize you with these parts too. Where diagrams are taken from other sources, you *must* cite the origin *in your figure caption*. For example:

**Figure 1:** Block diagram showing the parts of a typical UV-visible spectrophotometer (*from Stone, Instrumentation for Chemists (7)*)

Many students find it helpful to write the introduction – at least in bullet-point form – as part of their preparation for the experiment. If you do this, remember to revise it after completing the discussion section to ensure that you have included all the relevant information. In general, focus on the aspects of theory and instrumentation relevant to the set of experiments you are performing. Thus, you would *not* discuss monochromators or bandpass filters for a report on FTIR spectrometry; similarly, you would *not* describe the size exclusion separation mechanism for a report on reverse-phase HPLC. Make sure you mention both the advantages and limitations of the technique, along with key areas of application. If your experiments involve comparing specific methods for sample presentation or instrument calibration, make sure you describe the ways in which the techniques are similar and different.

Suppose, for example, that you were describing a set of experiments performed using UV-visible spectrophotometry. Your theoretical section would need to provide an overview of the interactions of light with matter, leading to a *short* presentation of Beer's Law and its limitations. Your instrumentation section would then provide a block diagram showing the layout of the key components in a spectrophotometer, highlighting the choice of light sources, the role of the monochromator and slits in obtaining useful spectra with minimal effects from polychromatic radiation, and the ordering of the source, wavelength selector, sample, and detector. You would also mention the limits of the technique for quantitative and qualitative analysis, such as type and phase of sample, concentration range, *etc.*

#### 5.2.4 Experimental (3-4 marks)

An experimental section should provide sufficient *relevant* information that: (a) it is clear how you obtained your results, and (b) another person could reproduce your experiment from the description provided without necessarily having exactly the same equipment. Experimental sections are usually sub-divided into: apparatus and chemicals used; general procedures; and procedures for specific experiments. When describing multiple experiments, sub-headings are often used so that each one can be referenced in the results and discussion easily.

When deciding what to include, consider the *purpose* of each step in the procedure, and whether using a different value for a parameter would significantly alter the outcome of the experiment. It is a good idea to assume that you are writing for someone who did *not* take the course, does *not* have a copy of the lab manual, and can only judge what you did by what you wrote.

- **Include** such things as operating temperature, target (nominal) concentration ranges, choice of wavelength for measurement, required sample pre-treatment, *etc.*

- **Exclude** detailed, step-by-step instructions for standard operating procedures, such as dispensing a solid by weight, filtering a precipitate, or accurately diluting a solution.
- **Include** the identity and version number of any software used to collect or process data is relevant
- **Exclude** the names of any local method files, or specific menu commands used within the software, are irrelevant.

Returning to our UV-visible example, the key information to provide would be that: the instrument was allowed to stabilise before use; adequate standards were prepared (list the *range* of concentrations, not individual values); a blank was run in order to correct all subsequent absorbance readings; standard spectra were obtained to identify suitable wavelengths for analysis; absorbance–concentration data was obtained (specify wavelengths, bandwidth, and pathlength used); and finally absorbance values were obtained for each sample. You would *not* need to describe the steps required to obtain this information using the specific spectrophotometer and computer software employed in your experiments, as this information will obviously vary between models and manufacturers. Similarly, you would want to identify the type of cuvette (pathlength and material) used to perform the measurements; you would *not* describe how to rinse, fill, and clean the cuvette.

### 5.2.5 Results (10 marks)

A results section provides a *written narrative account* of the results from each experiment, including a description of any steps required to convert raw data into the final form for subsequent analysis and discussion. For short articles describing a single experiment, the results may be combined with the discussion; for longer reports covering multiple experiments, however, it is more common to keep the results and discussion sections separate. In this case, sub-headings are used to distinguish different experiments, which are presented in the same order as in the experimental section.

Raw data is generally *not* included here, but may be attached in an appendix or supplied as supplementary data. Similarly, you would include carefully selected, *representative* chromatograms or spectra for your key findings, rather than every single one recorded during the course of the experiments. Data should be presented in either tabular or graphic form (whichever is more appropriate), but *rarely if ever* both. Always use the correct number of significant figures or decimal places to report measured values. Perform error analysis or statistical significance tests whenever appropriate.

Calibration data should normally be provided as a properly formatted and labelled graph, showing both the calibration points (with error bars) and the regression line. If errors in the calibration points are too small to be apparent on the graph, this should be indicated in the figure caption. Relevant regression statistics can either be included on the graph, or provided separately in a clearly labelled table. Sample concentrations (with the associated uncertainties) derived using the calibration curve should similarly be presented in tabular form, while the text describes how these values were derived. Additional data (such as measured responses, means, *etc.*) should be relegated to the appendix or supporting material, as appropriate.

To illustrate these points using our UV-visible example: “The background-corrected absorbance of each chromium sample was measured in triplicate, and the mean values used to determine the sample concentration and associated uncertainty ( $C_0 \pm s_{x0}$ ) using the regression

equation and associated statistical parameters.” Similarly, in describing a more complicated mathematical process for obtaining results: “The molar absorptivities at the wavelengths of maximum absorbance for the cobalt and chromium species were calculated from the calibration data obtained at each wavelength using Beer’s Law (Table 2). The concentrations of cobalt and chromium in the sample mixture was then calculated from the absorbance values at these same wavelengths by the method of simultaneous equations, yielding values of  $2.37 \pm 0.01$  and  $5.42 \pm 0.02$  mmol/L, respectively (see appendix 2 for details.)”

Finally, remember that *every* table and figure included in your report *must* have a caption clearly identifying what it is. Also be sure to *cite the source* for any diagram, figure, or photography reproduced from an article, book, or web site. (This applies equally to *all* sections of your report.)

- ➔ If you did **not** take the CHM217 introductory course, check with your lab instructor or TA as to what constitutes appropriate statistical analysis for each experiment!

### 5.2.6 Discussion (25 marks)

This is the *most important* section of a report or article, and you should invest your time in writing it accordingly. It is important to distinguish between results and discussion, even if these are combined in a single section: **The results section** presents the *observations* and values from your experiment, including the methodology for any data manipulation required to obtain these. **The discussion section** describes the *interpretation* of these results, particularly with respect to the theory or experimental hypothesis outlined in the introduction.

This includes what the results mean, what they prove or disprove (if anything), and any further implications of these findings. This is also the place to address any potential shortcomings in the experimental methodology, or to evaluate sources of error and uncertainty in the results; if an experiment was incomplete or failed, this is also the place to discuss what happened, why it happened, and how to avoid similar problems in the future.

For the cumulative reports expected in this course, you should not only discuss each experiment in terms of the results obtained, but also discuss what the experiments *collectively* demonstrate about the technique being used. For example, what kinds of samples and types of analysis is this instrument particularly suitable for? If the experiment exploited a particular calibration method or feature of the instrumentation, how successful was this? Why was it relevant to use for the particular sample being measured? How would you decide whether or not to use this technique for other samples?

You will find specific questions for each technique at the end of (a) each experiment and (b) each section in the lab manual (under “Elements for report discussion”): work the answers to **all** these questions into your discussion in the relevant place. They should form an integrated part of your discussion, rather than simply a list of answers: this is a *report*, and *not* an exam paper!

### 5.2.7 Conclusion (2 marks; maximum 250 words)

A conclusion provides both a summary of the preceding results and discussion, as well as a place to reflect on what can be learned from this information. It will obviously present some of the same information included in the abstract, but it should do more than simply duplicate this.

Summarise *what you learnt* about the technique (theory and practice), as well as the results and implications of any sample analyses performed during the set of experiments. This should include a *short* restatement of the principle points from your discussion.

### 5.2.8 Reference List

This should comprise a complete, sequentially numbered list of all sources and citations used in writing your report. On-line resources\* are limited to a *maximum* of 10% of your reference list; marks may be deducted for improper referencing or having too many websites as references.

\* This does *not* include published peer-reviewed journals that you accessed on-line; rather, it refers to company web sites, on-line encyclopaedias, *etc.*

### 5.2.9 Appendices

During the course of your experiments, you will most likely have collected a large number of original spectra or chromatograms, as well as all your calibration graphs and detailed calculations. It is not necessary, neither is it desirable, to include *everything* with your report. You should carefully select and attach *relevant* supplementary data or extended calculations that support your results and discussion; *don't* simply include every chromatogram, spectrum, or chart!

### 5.2.10 Overall Writing Assessment (5 marks):

The overall quality of your writing will be assessed with respect to logical flow, structure, clarity, and effectiveness. Writing well takes time, so plan your work schedule accordingly; do your calculations and write-up *as you perform each experiment*, rather than leaving it to the last minute to start your report. Spreading the work out in smaller chunks makes it much more manageable, and is a much more effective use of your time and energy, than trying to “pull an all-nighter.” You will also retain more of what you learn that way, which will be important for your tests and final exam.

## 5.3 Plagiarism & Proper Referencing

Plagiarism is the act of presenting the words and ideas of others as your own. One of the reasons for having you write these reports is to help you develop your communication, analysis, and critical thinking skills, as well as deepening your understanding of the techniques covered by the course. It is therefore essential that you should use your own words to demonstrate what you are learning.

All students are reminded of the University's policy on plagiarism. Specifically, you should use your own words in writing the introduction, discussion and conclusions of your report. Short quotations are admissible, but they must be clearly identified as such and include the appropriate citation or reference. Similarly, diagrams from texts and journals can be copied and pasted into your report, but you must include a full citation for the source either in the figure caption or as a footnote or reference to the caption.

Finally, when summarising information from several paragraphs in a source text, it is *not* sufficient to simply copy key sentences into a single paragraph with a reference number after each one; you should express the information in your own words, citing the source of the information only once. Consider the following examples, based on a section in the course text (compare with the original text as cited below the examples). You will find additional examples on the Writing Support Centre's web site.

**The following is *not* acceptable:**

The energy level diagram for the outer electrons of an element provides a convenient method for describing the various methods of atomic spectroscopy (Fig. 1).<sup>1</sup> The energies of atomic orbitals are indicated on the diagram by horizontal lines.<sup>1</sup> Note that the *p* orbitals are split into two levels that differ slightly in energy.<sup>1</sup> Similar differences exist in the *d* and *f* orbitals, but their magnitudes are ordinarily so slight as to be undetectable.<sup>1</sup>

1.) Skoog, Holler, & Nieman, "Principles of Instrumental Analysis" (5th ed.), Saunders College Publishing, Philadelphia 1992, pp. 192–193

**Note:** Using a citation on every copied sentence does *not* make the plagiarism acceptable.

**The following is *not* acceptable:**

The energy level diagram for the outer electrons of an element provides a convenient method for describing the various methods of atomic spectroscopy (Fig. 1). The energies of atomic orbitals are indicated on the diagram by horizontal lines. Note that the *p* orbitals are split into two levels that differ slightly in energy. Similar differences exist in the *d* and *f* orbitals, but their magnitudes are ordinarily so slight as to be undetectable.

**Note:** You should *never* copy text without providing a citation and indicating that you are directly quoting the source.

**The following *is* acceptable:**

Figure 1 shows an energy level diagram for the outer electrons of atomic sodium. Such diagrams, in which horizontal lines represent atomic orbitals, are convenient for describing the different forms of atomic spectroscopy. Note that the orbitals within each *p*, *d*, and *f* subshell are split into levels differing only slightly in energy rather than being fully degenerate. This difference in energy is ordinarily so small for orbitals in the *d* and *f* subshells that it cannot be detected.<sup>1</sup>

<sup>1</sup>) Skoog, Holler, & Nieman, "Principles of Instrumental Analysis" (5th ed.), Saunders College Publishing, Philadelphia 1992, pp. 192–193.

**The following *is* acceptable:**

Figure 1 shows an energy level diagram for the outer electrons of atomic sodium. As explained by Skoog *et al*<sup>1</sup> such diagrams, in which horizontal lines represent atomic orbitals, are convenient for describing the different forms of atomic spectroscopy. Note that the orbitals within each *p*, *d*, and *f* subshell are split into levels differing only slightly in energy rather than being fully degenerate. This difference in energy is ordinarily so small for orbitals in the *d* and *f* subshells that it cannot be detected.

<sup>1</sup>) Skoog, Holler, & Nieman, "Principles of Instrumental Analysis" (5th ed.), Saunders College Publishing, Philadelphia 1992, pp. 192–193.

## 5.4 Getting Help

Ask questions about the experiments and your reports *during the lab* – unfortunately, it is not possible to support separate office hours for your lab demonstrators. Be sure to *take notes* when discussing work with your TA! You can also talk to the lab instructor during *any* lab period, or make an appointment by email. See also: <http://www.writing.utoronto.ca/>

## 5.5 Weekly Executive Summary

The purpose of an executive summary is to communicate as simply and clearly as possible the essential facts and conclusions derived from a research project, experiment, or analysis. It is essentially a combination of abstract and conclusion, and is typically no more than a single page in length. An effective executive summary should allow the person reading it to take immediate action based on the findings without necessarily having to read the entire report.

In order to prepare your executive summary, you should first complete all calculations and data analysis required for the experiment you have just completed. Review this information together with the goals of the experiment, making sure you have completed all the steps in the post-lab section of the experimental procedure. Be sure to include copies of any graphs, along with the results of any statistical analysis, in your laboratory notebook when you do this.

You can now write up your executive summary. It should state simply and clearly what you did, why and how (in general terms) you did it, what you found, and what you can conclude from this information. If part of the experiment was unsuccessful, you should include this information, your conclusion as to what went wrong, and what you would recommend in order to avoid this issue in the future.

Type your executive summary (include your name, demo group, and the number and title of the experiment) in 150-250 words. Print out two copies: submit one to your demonstrator, and attach the other to your lab notebook. Each summary will be graded out of four marks, depending on how well the summary meets the criteria given above.

If you have any questions or concerns about writing your executive summary, please contact Professor Stone: you can either drop by during one of the lab periods, or make an appointment to meet outside of lab hours.

## 6. LABORATORY SAFETY

By its very nature, experimental chemistry is potentially dangerous. Although every effort is made to identify and (where possible) eliminate risks, it is not possible to anticipate every hazard. Ultimately *you, and you alone*, bear the responsibility for your own personal safety in the laboratory. You must therefore take the time to read and understand all the safety information and procedures outlined in this section. While the amount of material may seem overwhelming, please realise that most of it is basic common sense. In fact, the majority of accidents are completely avoidable, and often result from a lack of preparation, being in a hurry, or simply not paying attention to what is going on.

### 6.1 Rules for Lab Safety

As part of your pre-lab preparation for each experiment, you should look up and include in tabular format in your lab notebook the toxicity and hazard information for *all* the chemicals you will be using. You should also review any safety precautions, emergency procedures, and chemical waste disposal instructions appropriate for the experiment(s) in question. When looking for this information, you need to think about both the physical form the substance will be in (*e.g.* gas, liquid, solution) and the amount you will be using (*e.g.* concentration). For example, it is much more important to wear gloves when handling concentrated rather than dilute (*e.g.* < 0.1 M) acid, but those gloves need to provide adequate protection. When deciding on what constitutes relevant safety procedures, consider aspects such as flammability, volatility, toxicity, *etc.* Should you be using the material only in a fume hood, for example?

**Work mindfully and systematically**  
**Exercise common sense at all times**  
**Always read the label on the bottle**  
**In case of chemical spills: wash immediately**  
**In case of fire: get out**  
**In case of inadequate preparation: stay home**  
**Dispose of all chemical waste properly – not down the sink**  
**Get a good night's sleep and eat a proper breakfast**  
**Always be aware of where you are and what you are doing**

### 6.2 Lab Dress Code

Laboratory attire must be protective from head to toe, and easily removed in the event of chemical spills. In particular, you should observe the following precautions:

- University policy requires that splash-resistant safety goggles *must* be worn at all times in the laboratory, even over prescription glasses, to protect against chemical splashes or flying objects. Safety goggles must be indirectly vented or non-vented, and comply with CSA Standard Z94.3-02. Please see <http://www.utoronto.ca/safety/documents/EyeandFacewearStd2004.pdf>
- You are advised to *not* wear contact lenses: vapours, liquids and foreign objects can become trapped behind them.

- Wear a full-length lab coat over your clothes, and keep it done up at all times in the laboratory. **University policy requires** that only 100% cotton lab coats may be used in a chemical laboratory. This is because synthetic fibres pose a much greater hazard in the event of fire or solvent spills.
- Wear long pants and fully enclosed, durable footwear – remember that spilled chemicals will tend to flow down towards any exposed toes or feet!
- Long hair should be tied back out of the way. Loose articles of clothing should also either be removed or secured so that they cannot fall into your experiment!
- Wear appropriate protective gloves *when necessary*, especially if you have any cuts or scrapes on your hands. Gloves should be of the correct type *for the specific chemical(s) you will be using* (see below).

Students are advised to keep a change of clothing handy, in case of an accidental spill or other incident.

### 6.3 Gloves

Gloves can protect your hands against accidental chemical contamination, but can also interfere with your ability to perform certain procedures such as pipetting. There are different types of gloves for different chemical applications. Disposable gloves, for example, are satisfactory for handling dilute aqueous solutions, but will **not** protect against strong acids, bases, and many organic solvents. Gloves can also be a source of chemical contamination! Disposable gloves should be made of material with an average thickness of **at least** 6 mil (0.006”).

- Wear appropriate gloves whenever handling concentrated acids and bases or organic solvents, or any substance that is harmful through skin contact. Thin disposable vinyl, latex, and nitrile gloves do **not** provide adequate protection against more than brief contact with such substances.
- Wear gloves of the appropriate *size*: they should not be loose (to avoid snagging); neither should they be stretched tight, as this thins out the glove and reduces dexterity.
- Thick nitrile or neoprene-coated, chemically-resistant gloves are recommended. These can be purchased from most hardware stores.
- **You must remove your gloves before:**
  - \* Handling any computers or instrumentation
  - \* Touching hair, skin, or clothing
  - \* Leaving the laboratory for **any** reason

Exception: if you are carrying samples or solutions, you can wear gloves, but you **must** have someone else open the doors for you.

Failure to do so can result in chemical transfer and potential personal injury. Students who ignore these instructions *will* have performance marks deducted, and *may* be asked to leave the laboratory, resulting in a zero mark.

- Used gloves are considered as contaminated waste, and should be disposed of in the green-coloured pails – do *not* throw them into the grey waste bins.

#### 6.4 Fire Precautions

When using flammable solvents (*e.g.* ethanol, acetone, *etc.*) do not light matches or Bunsen burners, and do not keep excess solvent in open beakers. Familiarise yourself with the location of all fire equipment, including the location of the fire extinguishers, safety showers, fire alarms, and emergency exits. In the event of a fire:

- If the fire is contained in a flask or beaker, cover the vessel to exclude oxygen.
- If spilled organic solvent is burning on the floor or bench top, *etc.*, sound the alert and leave the lab without being asked to do so.
- If someone near you is on fire, smother the flames with a lab coat or fire blanket. If the safety shower is nearby, douse the victim. Do not wait for someone else to act.

#### 6.5 General Precautions

As mentioned above, you should familiarise yourself with the properties of any chemical substances that you will be working with before coming to the lab. These should be recorded in your lab notebook. Useful sources of information include the Merck Index and the NIOSH/OSHA Pocket Guide to Chemical Hazards. Additional sources of information can be found from the Health & Safety section of the Chemistry web site (<http://www.chem.utoronto.ca/>). On-line access to the CRC Handbook of Chemistry and Physics and the NIOSH Pocket guide, together with other resources, is available via the links on the Chemistry library web. Additional links to chemical and safety information web sites can be found at the end of this section or on the course web site.

- Smoking, eating and drinking are strictly prohibited in the laboratory.
- Handle all chemicals with care, using gloves where appropriate. *Always* read the label on a container to make sure it is both the right chemical *and* the right concentration.
- ☠ Thin, disposable gloves are *not* appropriate for use with halogenated solvents such as chloroform; they have been known to leak on occasion.
- When dispensing chemicals:
  - *always* hold a container by the label to minimize accidental contact
  - *always* clean up chemical spills *immediately*; *never* leave a mess behind
  - *always* return chemicals *promptly* to the correct location
  - *always* make sure container lids are on properly
- Remove your gloves before using instruments and computers, or exiting the lab, in order to avoid possible chemical contamination.

- Make sure you have read, and understood, the correct procedures to follow for each operation such as weighing, making solutions, or filtering. Use of the correct procedure not only minimizes the risk of personal injury, but also ensures accurate, precise, and reliable data.
- Splashes of corrosive or toxic substances should be washed immediately from the skin or eyes with copious amounts of cold water. If a toxic or corrosive substance splashes your lab coat, take it off before the contamination can soak through to your clothing.
- Clean up any chemical spills immediately. Neutralise strong acids with bicarbonate, then wash with water. Soak up organic solvents using a cloth or towel, leaving it in a fume hood until the solvent evaporates.
- Report any broken thermometers or glassware to lab staff *immediately*.
- **Always** use the fume hood when instructed to do so. **Never** open the fume hood door beyond the limits clearly marked on the side frames.
- **Always** make sure your chemicals are placed inside the fume hood, and *not* perched right at the edge of the hood.
- Do *not* dispose of chemicals or broken glassware in the garbage bins; use the appropriate, labelled container for all chemical waste and place any broken glass in the white bins. *Always* check the label on the container before emptying your waste into it.
- To avoid cuts, *always* handle glassware *gently*. If you break any glassware, ask for the broom and dustpan and sweep up the pieces as soon as possible. If you cut yourself on a piece of glassware, wash the cut immediately with cold water to avoid chemical contamination and apply pressure to stop further bleeding. Seek further aid from the prep room

**ALL injuries, spills, or breakages, no matter how slight, MUST be reported to the laboratory staff as soon as possible. In the event of personal injury, an accident report form must be completed and submitted to the departmental health and safety representative.**

## 7. Disposal of Chemical, Glass, Paper, and Plastic Waste

It is imperative that *everyone* in the lab takes steps jointly to (a) minimise the amount of chemical and other waste generated (environmental responsibility) and (b) dispose of any waste correctly (legal responsibility). In particular, students should note that it is ***both illegal and potentially hazardous to pour waste chemicals down laboratory sinks***. Waste containers are provided for each specific experiment, either in one of the fume hoods or at the end of the bench. In addition, designated containers are provided throughout the room for different types of waste.

Please observe the following at all times:

- Broken glass that is ***not*** chemically contaminated (*i.e.* was clean when broken, or has been rinsed after breaking) can be placed in the **teal**-coloured wheeled bins; glass in these bins is collected and sent for recycling.

- Clean plastic waste, such as cracked plastic wash bottles, should be placed in the **orange**-coloured wheeled bins for recycling.
- Any contaminated glass, paper, or plastic **must** be disposed of in the green pails with the white lids.
- Disposable gloves are also considered as contaminated waste, and **must** be disposed of in the green pails.
- General paper waste can be placed in the large grey bins



*Waste containers for recycling clean glass (left) and plastic (right)*

- There are separate, labelled, solid and aqueous chemical waste containers provided either on the bench or in one of the fume hoods for each experiment.
- Any solution containing a halogenated organic substance (such as dichloromethane or bromobenzene) **must** be disposed of in the halogenated waste container in the south fume hood in LM6.
- Non-halogenated, flammable, hydrophobic organic waste (such as ethyl acetate, toluene, *etc.*) should be disposed of in the large red waste can beside the south fume hood in LM6.
- Any aqueous solutions or flammable, hydrophilic organic waste should be disposed of in the appropriate labelled waste bottle.

## 8. Cleaning Glassware for Analytical Chemistry

Contaminated glassware is a very common source of gross error in analytical chemistry, especially when trying to identify and measure substances present at low concentrations. Most breakages – and the risk of accident – also occur during cleaning. It is therefore important that

you use correct protocols when cleaning analytical glassware. The following general procedures will help you avoid typical mistakes; detailed procedures for specific items are included with each experiment where appropriate. Note that **you** are responsible for cleaning and returning any glassware (along with any chemicals and equipment) you use to the correct location by the end of the lab period.

- Glass is slippery when wet!
  - Most people know this very well, but seem to forget it when in the lab. Please do **not** shake **any** glassware to remove water from it, especially if you happen to be wearing disposable gloves. Gravity and evaporation both work nicely, if you give them a chance.
- Paper towel is **not** contaminant free!
  - In addition to surface chemicals, paper towel tends to leave small fibres behind that can easily clog burettes and pipettes. Use it only to wipe down the *outside* of glassware; let glassware drain after rinsing, then dry naturally.
- Compressed air is **not** contaminant free!
  - There is a small amount of pump oil present in the compressed air supply at the fume hoods, which leaves glass greasy. Additionally, the high pressure can cause objects to fly considerable distances...
- Wash bottles are for washing!
  - Volumetric flasks, pipettes, and burettes should never be held under a tap. Drain any remaining chemicals into a beaker for subsequent disposal, and then use a continuous stream of distilled or deionized water from a wash bottle to ensure the glass is clean. Please also note that the distilled water pressure at some sinks is *very high*, and can easily knock a beaker clean out of your gloved hand...
- Balancing acts do **not** belong in the laboratory!
  - Volumetric flasks are inherently unstable when balanced upside down, not to mention costly to replace when they then fall over. Flasks with wide enough necks can be slid onto a peg on the drying racks over the sinks. Otherwise, just leave the stopper out of the flask and let it dry naturally. If you need to use it for another solution, just rinse it with that solution first.
- Ethanol is actually a chemical!
  - When removing ink from the outside of plastic bottles and glassware, spray a **small** amount of ethanol onto a piece of paper towel, and then wipe the surface clean. You may dispose of the towel in the grey bins. Allowing the ethanol to enter the drain is actually illegal in a laboratory (although pouring beer and wine into the sink at home is a different sort of crime...)

## 9. Computer Tutorials

Four of the techniques covered in this course are the subject of computer-assisted tutorials, which are available on a number of the computers in Analest (LM 9). You may access these tutorials whenever the computers are free; please check with Dan Mathers regarding an appropriate time (generally Monday to Friday, 9 am to 5 pm, when not required by other courses.)

## 10. Reference Materials

The following may prove useful when preparing for labs and writing your reports. Many of the texts are available from the Chemistry and Gerstein libraries.

### General Analytical Chemistry Texts:

- “Analytical Chemistry” by G. D. Christian, Wiley
- “Quantitative Chemical Analysis” by D. C. Harris, W. H. Freeman
- “Modern Analytical Chemistry” by D. Harvey, McGraw-Hill
- “Fundamentals of Analytical Chemistry” (8th ed) by Skoog, West, Holler and Crouch, Saunders College Publishing
- “Principles of Instrumental Analysis” (6th ed) by Skoog, Holler & Nieman, Saunders College Publishing (CHM 317 text)
- “Data Analysis for Chemistry” by D. B. Hibbert & J. J. Gooding, Oxford University Press

### Reference Materials:

- CRC Handbook of Chemistry and Physics
- The Merck Index
- Vogel’s “Quantitative Inorganic Analysis”, Longmans
- “Official Methods of Analysis of the AOAC”, ed. K. Helrich, VA:AOAC International
- “Encyclopedia of Analytical Chemistry”, ed. R. A. Meyers, Wiley

### Primary Journals:

- Analyst
- Analytical Chemistry
- Analytica Chimica Acta
- Analytical Biochemistry
- Clinical Chemistry

### Chemical Information sites:

- MSDS: <http://ccinfoweb.ccohs.ca/msds/search.html>
- MSDS: <http://msds.ehs.cornell.edu/msdssrch.asp>
- MSDS: <http://physchem.ox.ac.uk/MSDS/>
- Safety: <http://ccinfoweb.ccohs.ca/cheminfo/search.html>
- Safety: <http://www.epa.gov/enviro/html/emci/chemref/>
- Safety: <http://atsdr1.atsdr.cdc.gov/toxfaq.html>
- NIOSH pocket guide: <http://www.cdc.gov/niosh/npg/>
- NIST Web Book: <http://webbook.nist.gov/chemistry/>

## 11. Your Laboratory Notebook

Good record-keeping is an essential part of any experimental science. Developing careful and precise habits regarding your lab notebook is therefore an important part of your chemical education, a fact recognised in the marking scheme. The following directions are to be followed at all times:

1. Use a hard-cover, bound, 8.5" × 11" notebook with ruled pages.
  2. Write your name, course, and demo group number on the outside cover and inside cover page.
  3. Number each page **in ink** on the top corner for easy identification. **Never** remove a page from your notebook, even if you have made an error; simply rule a single diagonal line through the relevant material.
  4. Reserve the first two or three pages for an index, and make sure you keep this up-to-date.
  5. Date every page as you work. If the work recorded on a page is spread over more than one day, record the dates in the margin(s).
  6. Always write in ink (preferably water-insoluble), **never** in pencil. If you make an error, cross it out by ruling a single line through it; do **not** use white-out, and do not tear out or cut the page.
- ☞ In certain jobs, this is a legal requirement to prevent the possibility of data fraud, so this is a good habit to develop.
- ➔ Marks will be deducted for failure to comply with these rules.
7. As you prepare for your next experiment, make any notes and calculations directly in your notebook so that you have them when needed. You do not need to duplicate the entire procedure step-by-step from the lab manual. Instead, make sure that you have a summary

of the key points of the procedure, including any specific weights, volumes, or concentrations, together with a reference to the relevant lab manual pages.

Example:

- prepare  $\sim 1.00$  M stock solution of sodium chloride by accurate dilution of  $\sim 5.84$  g to 100.0 mL with deionised water
  - prepare 100 mL each of sodium chloride standards  $1.00 \times 10^{-1}$  –  $1.00 \times 10^{-6}$  M by serial dilution with deionised water
  - calibrate sodium ISE using 20.00 mL standard + 2.00 mL ISA
  - analyse samples by Na ISE using 20.00 mL sample + 2.00 mL ISA
  - see lab manual pp. 36–38 for measurement protocol
8. Record all your weights, readings and results directly in your notebook as you perform the experiment. Include notes of any particular problems or difficulties encountered with samples or procedures. Also include any calculations, e.g. formula weights, mass required for specific concentrations, unit conversions, final results, error propagation, etc.
    - ➔ Marks will be deducted for not recording data in your notebook as you perform the experiment. You should never rely on memory! For experiments where the data is collected and recorded on a computer, always keep a record of which file names are associated with which sample or standard, together with the order the measurements were performed (see below). Not only is this good laboratory practice, it will make your life much easier should you need to check the data!
  9. Write on only one side of each page. Use clear headings throughout your work so that you can find information easily and quickly when you come to prepare your reports. Write legibly, and space out your results and tables so that they are clear and easy to find. Use underlining to highlight final results and conclusions. Use the blank facing page as a place to attach graphs or show calculations performed post-lab in working up your data.
  10. Where results must be processed by computer or obtained from a computer printout, always glue or tape a copy into your notebook at an appropriate location – leave a blank page for this if necessary. The same applies for results obtained by your partner during joint experiments: photocopy the relevant pages from each notebook, and then glue the copy into your book in the appropriate place.
  11. Keep a running record in your notebook of the order in which you do the measurements, and the corresponding file names where the results have been saved. Always label computer printouts immediately with your name, the identity of the sample/standard, and the corresponding page in your lab notebook.
  12. Your demonstrator will check your lab notebook periodically throughout each period. You **must** also have him or her validate your notebook at the end of each experiment **before** you can leave the lab. You will lose marks if this is not done, so don't forget this important step in your hurry to leave!

- ➔ You will lose marks if you leave without having your notebook validated. GLP and statutory compliance regulations requires your records to be corroborated by a supervisor.
- 13. When analysing your data in preparation for making your report, record all your calculations and conclusions in your lab book. Your demonstrator will be checking that you have done this at the start of your next lab.
- ➔ You will lose marks if have not recorded your final calculations in your notebook. GLP and statutory compliance regulations requires a record of exactly how you reached your conclusions in respect to any measurement.

## 12. Significant Figures, Decimal Places, and Error Propagation

In any practical science, all calculations and results should take the errors and uncertainties associated with actual measurements into account. In this context, the **error**  $E_x$  in a measured value  $x$  is defined as the *actual* difference between the true and measured values:  $E_x = x_{true} - x$ . While such measurement errors *always* exist, it is not always possible to determine them since the true value may not be known. In most cases the best we can do is provide an *estimate* of the error, which we term the **uncertainty**; any value  $x$  therefore always has an associated uncertainty  $\pm\Delta x$ , which must be accounted for in any subsequent calculations. The uncertainty  $\Delta x$  may be due to known sources of error, random fluctuations inherent in instrumental operation, or the limiting resolution of a readout or display. Uncertainty may be expressed as an actual error (when known), an estimated error, a standard deviation, or confidence interval. Uncertainty also exists in any quantity derived from one or more measured values, and must be determined by a procedure known as **error propagation**.

### 12.1 Determining the uncertainty, $\Delta x$ :

- Derived or measured values always have an **implied uncertainty** if none is stated, which is taken as  $\pm 1$  in the last significant figure. Uncertainties *always* have the same units as the value for which they provide an error estimate. For example:

$$M_m = 242.13 \pm 0.01 \text{ g/mol} \quad E = -163 \pm 1 \text{ mV}$$

$$A = 0.137 \pm 0.001 \text{ AUC} = 1.00 \times 10^{-3} \pm 1 \times 10^{-5} \text{ mol/L}$$

- When the arithmetic mean of a set of  $n$  repeated values is calculated, the uncertainty is based on the unbiased standard deviation,  $s$ . This is denoted on most calculators as either  $\sigma_{n-1}$  or  $s_{n-1}$  and is calculated as:

$$s = \sqrt{\frac{\sum_{i=1}^n (x_i - \bar{x})^2}{n-1}}$$

👉 Save time and avoid simple mistakes – use your calculator for this!

Note that a standard deviation, being an estimate of the error in the mean value, only has *one* significant figure; it is customary to report the first non-significant digit also, in order to avoid premature rounding in subsequent calculations. For example:

**Example 1:**  $m = 10.0120, 10.0051, 10.0073, 10.0046, 10.0111$  g

$$\bar{m} = 10.0162_8 \text{ g}, s = 0.01_7 \text{ g}, n = 5$$

$$\therefore \bar{m} = 10.02 \pm 0.02 \text{ g } (\bar{m} \pm s, n = 5)$$

Note that, in this example, the mean value actually has *fewer* significant figures than the individual readings as a direct consequence of the value of the standard deviation!

- Sometimes, an instrument is incapable of resolving the inherent variation between repeated measurements, resulting in a string of identical values with  $s = 0$ . In this case, the uncertainty is expressed as  $\pm 1$  in the last figure of the instrument display.

**Example 2:**  $E = 107, 107, 107$  mV

$$\bar{E} = 107 \text{ mV}, s = 0 \text{ mV}, n = 3$$

$$\therefore \bar{E} = 107 \pm 1 \text{ mV } (n = 3)$$

## 12.2 Error propagation with implicit uncertainties:

When performing calculations using values that have an implied uncertainty, it is essential to observe the correct rules for determining the uncertainty  $\Delta q$  in the final value,  $q$ , as this will determine the correct number of significant figures to which the result may be quoted.

- When **adding or subtracting**, the least significant digit in the result is determined by the value with the **fewest decimal places**.
- When **multiplying or dividing**, the least significant digit in the result is determined by the value with the **fewest significant figures**.
- When a calculation involves both types of operation (additive and multiplicative), observe the order of mathematical precedence and determine the correct numbers of significant figures for intermediate values.
- It is **always** advisable to retain *at least* the first non-significant digit in intermediate values in order to prevent premature rounding of the final result. Non-significant digits should be clearly denoted using either subscripting, underlining, or enclosing them in parentheses.

**Example 3:** calculate the molar mass of calcium carbonate

$$1 \times 40.08 = 40.08$$

$$1 \times 12.011 = 12.011$$

$$3 \times 15.9994 = 47.9982$$

$$= 100.0892 \quad \therefore M_m(\text{CaCO}_3) = 100.09 \text{ g/mol}$$

Notice how, in this example, the molar mass *correctly* has 5 significant figures, even though the atomic mass of calcium was only known to four significant figures! This is because the *largest* uncertainty in any of the masses is  $\pm 0.01$  g/mol, so the error in the sum total lies in the second decimal place.

**Example 4:** calculate the mass of calcium carbonate required to make 100.0 mL of an 0.200 M solution:

$$C = \frac{m \text{ (g)}}{M_m \text{ (g/mol)} \times V \text{ (L)}} \therefore m = C \times M_m \times V$$

$$\therefore m = 0.200 \frac{\text{mol}}{\text{L}} \times \frac{100.0 \text{ mL}}{1000 \text{ mL/L}} \times 100.08_{92} \frac{\text{g}}{\text{mol}}$$

$$\therefore m = 2.00_{18} \text{ g} = 2.00 \text{ g of calcium carbonate}$$

Notice how, in this example, the final result can only be given to 3 significant figures, even though the volume and molar mass are known to 4 and 5 significant figures, respectively. (Remember that leading zeroes before the decimal place are *not* counted towards the number of significant figures; we could clarify this by writing  $C = 2.00 \times 10^{-1}$  M.)

- When calculating logs and antilogs, remember that the number of significant figures associated with a value  $x$  determine the number of decimal places reported for  $\log x$ . Similarly, the number of decimal places for a value  $y$  determines the number of significant figures reported for  $10^y$ .

### 12.3 Error propagation with calculated uncertainties:

In analytical and physical chemistry, it is important to explicitly calculate the effect of individual uncertainties on the overall uncertainty in the final result. In fact, the rules in the preceding section are based on the mathematical procedures one must use to do this. It is therefore important to evaluate the uncertainties in additive and multiplicative steps independently, as different rules apply.

- When adding or subtracting values with random errors, *e.g.* if  $q = x + y$  or  $q = x - y$  then the uncertainty  $\Delta q$  in  $q$  is:

$$\Delta q = \pm \sqrt{(\Delta x)^2 + (\Delta y)^2}$$

**Example 5:** the uncertainty in the difference between two weights, each measured to the nearest  $\pm 0.0001$  g:

$$m = m_{\text{final}} - m_{\text{initial}} = 23.2476 - 21.1942 \text{ g} = 2.0534 \text{ g}$$

$$\Delta m = \sqrt{(0.0001)^2 + (0.0001)^2} = \pm 0.00014 \text{ g}$$

- When multiplying or dividing values with random errors, e.g. if  $q = ax \times y$  where  $a$  is a constant, the uncertainty  $\Delta q$  in  $q$  is:

$$\Delta q = \pm q \sqrt{\left(\frac{\Delta x}{x}\right)^2 + \left(\frac{\Delta y}{y}\right)^2}$$

**Example 6:** the uncertainty in the concentration obtained by dissolving 2.0534 g of  $\text{CaCO}_3$  in water and making to volume in a 100.0 mL volumetric flask

- The uncertainty in the mass from above is  $\pm 0.00014 \text{ g}$
- The molar mass from above is  $100.09 \pm 0.01 \text{ g/mol}$
- The uncertainty in flask volume is  $\pm 0.08 \text{ mL}$

$$C = \frac{m}{M_m \times V} = \frac{2.0543 \text{ g}}{100.08_{92} \text{ g/mol} \times 100.0 \text{ mL}} \times \frac{1000 \text{ mL/L}}{100.0 \text{ mL}} = 2.05157 \times 10^{-1} \text{ mol/L}$$

$$\Delta C = \pm C \sqrt{\left(\frac{\Delta m}{m}\right)^2 + \left(\frac{\Delta V}{V}\right)^2 + \left(\frac{\Delta M_m}{M_m}\right)^2}$$

$$\therefore \Delta C = \pm 0.205157 \sqrt{\left(\frac{0.00014}{2.0534}\right)^2 + \left(\frac{0.08}{100.0}\right)^2 + \left(\frac{0.01}{100.08_{92}}\right)^2}$$

$$\therefore \Delta C = \pm 1.66 \times 10^{-4} \text{ M} \quad \therefore C(\text{CaCO}_3) = 0.2051_6 \pm 0.00017 \text{ mol/L}$$

## 12.4 Rounding values from calculations:

Whether dealing with implied or actual uncertainties, it is essential to round the final value correctly. Most textbooks describe the procedure adopted in computer science, and commonly implemented on calculators, where values are rounded up if the first non-significant digit is greater than or equal to 5, but rounded down if less than 5. Unfortunately, this leads to some bias in the results for cases where the non-significant portion of the number is exactly 5. In analytical chemistry, we therefore implement a third rule: if the non-significant portion of the number is exactly 5, the result is rounded to the *nearest even number*. This means that half the time the result will be rounded *up*, and half the time it will be rounded *down*.

- In computer programming, this is sometimes referred to as “banker’s rounding”; if the bank always rounded the interest on your loan up, you would be overcharged!

### Example 7:

- $q = 13.379_{512}$  – the non-significant digits are  $> 0.000_5$  so we round **up**
- $q = 227.63_{439}$  – the non-significant digits are  $< 0.00_5$  so we round **down**
- $q = 1.141_5$  – the non-significant digits are  $= 0.000_5$ ; round to 1.142
- $q = 10.76_5$  – the non-significant digits are  $= 0.00_5$ ; round to 10.76



### 12.5 Outliers, Confidence Intervals, and Significance Tests:

- If you suspect that one or more values in a set of replicate measurements is an outlier, apply Grubb's test to the data to see if the suspect value can be rejected (see lab manual or the course text)
- When a final result is the average of several individual determinations, calculate the 95% confidence interval about the mean using the  $t$ -statistic (see lab manual or course text)
- If an experiment calls for either a comparison of the measured mean with a known true value (test of accuracy and bias) or the comparison of the standard deviations of two sets of results (test of precision), use the appropriate statistical test ( $t$ -test or  $F$ -test, respectively)

### 13. Calibration, Regression, Interpolation, and Bracketing

Most instrumental techniques require proper calibration of the instrument for accurate quantitative analysis. This is covered in detail in the course text. In addition, the course web site contains links to a self-guided tutorial on the use of Microsoft® Excel™ to perform linear regression analysis, and the correct calculation of the estimated errors in values derived from such calibration curves by either interpolation or extrapolation. This section provides a brief overview of the key points.

- Concentration is always the independent ( $x$ -axis) variable
- The number of significant figures for each axis scale is determined by the uncertainty in your standard concentrations and measured readings
- Always include a title, axis labels (with correct units), and a legend. The title should refer to the *experiment*, not simply "A plot of  $y$  versus  $x$ ". So, you might have a graph titled, "Calibration Curve for the Determination of Cobalt by Absorbance at 515 nm"
- Perform your own regression analysis on your calibration data, even if the instrument or software does this for you
- *Never* force a regression line through zero (the origin); you should, however, include the blank reading if available.
- *Always* calculate the slope, intercept, standard error of the regression, and the correlation coefficient ( $r$  or  $r^2$ ) for a calibration curve. You should also calculate the limit-of-detection from the regression data.
- For replicate measurements, *either* plot them as discrete values for each concentration *or* as mean values with  $y$ -error bars to indicate  $\pm 1$  standard deviation for each standard. Clearly indicate if you have done the latter but the error bars are too small to be visible on the plot.

Note that the use of Excel™ to perform many of the above is described in the on-line tutorial on calibration and regression, complete with numerous examples and sample files that you may

download. If you are unfamiliar with Excel, or any of the available statistics programs such as MAPLE, MiniTab, *etc.*, you should spend some time working through this material; you will waste a lot of time writing up lab reports if you do all the calculations by hand! See:

<http://www.chem.utoronto.ca/coursenotes/analsci/stats/>

The figure on the next page summarizes the key points of calibration, regression, and the limit of detection. In order to fit a linear calibration function  $y = bx + a$  to the experimental data, we use regression analysis (the mean values here are the mean  $x$  and  $y$  values for the calibration data):

Calculate the slope: 
$$b = \frac{\sum_i \{(x_i - \bar{x})(y_i - \bar{y})\}}{\sum_i (x_i - \bar{x})^2}$$

Calculate the intercept:  $a = \bar{y} - b\bar{x}$

Calculate the correlation coefficient:

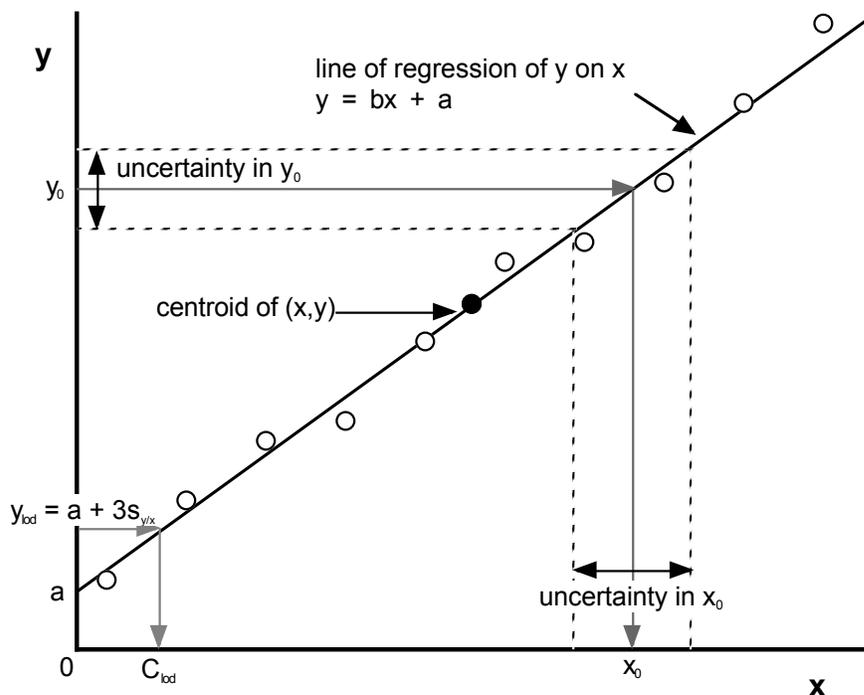
$$r = \frac{\sum_i \{(x_i - \bar{x})(y_i - \bar{y})\}}{\left\{ \left[ \sum_i (x_i - \bar{x})^2 \right] \left[ \sum_i (y_i - \bar{y})^2 \right] \right\}^{1/2}}$$

Calculate the standard error of regression: 
$$s_{y/x} = \sqrt{\frac{\sum_i (y_i - \hat{y}_i)^2}{n - 2}}$$

Note that many of these values can be calculated in Excel easily using the built-in LINEST function. For details, please see the on-line stats tutorial page on errors in regression analysis (<http://www.chem.utoronto.ca/coursenotes/analsci/stats/ErrRegr.html>).

### 13.1 Limit-of-Detection:

For linear response–concentration calibration plots, you should always calculate the limit-of-detection for your method and measurement conditions. In chemical measurement, this is defined as *the concentration giving the smallest detectable signal above the baseline (background) noise*. Technically, this should be calculated based on repeat measurement of a blank (sample matrix with *no* analyte present). Practically, this is often unfeasible due to time constraints. A common alternative is to use the calibration data and regression line, as shown:



*Line of regression of  $y$  (instrument response) on  $x$  (concentration) using least-squares, showing the concentration limit-of-detection ( $lod$ ) and the determination of an unknown concentration  $x_0$  from a measured response  $y_0$*

To calculate the limit-of-detection:

1. Determine the smallest detectable signal,  $y_{lod}$ , as *either*:
  - the blank plus three times the standard deviation of the blank ( $n > 20$ )

$$y_{lod} = y_{blank} + 3s_{blank}$$

- the intercept,  $a$ , plus three times the standard error of the regression

$$y_{lod} = a + 3s_{y/x}$$

2. Convert the value of  $y_{lod}$  into the corresponding concentration limit-of-detection using the calibration equation from the regression analysis:

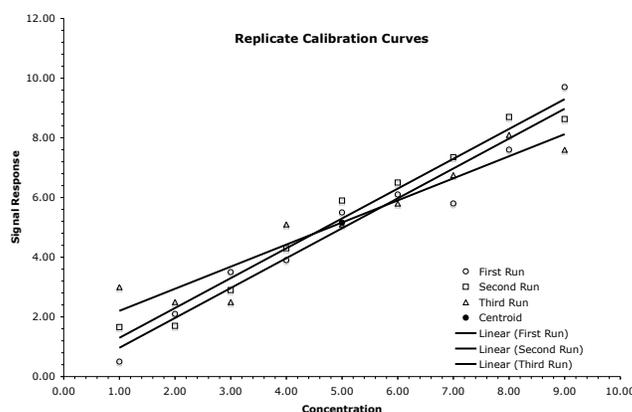
$$C_{lod} = \frac{y_{lod} - a}{b}$$

Note that, when using the regression line, steps 1 and 2 can be combined to yield a simplified form of the equation:

$$C_{lod} = \frac{3s_{y/x}}{b}$$

### 13.2 Interpolating a Value:

In order to calculate the concentration of a sample from its measured response, you will need to make an interpolation of the response,  $y_0$ , using the response equation derived from your regression analysis. The uncertainty in the resulting concentration,  $x_0$ , can be calculated from the regression statistics as demonstrated in the on-line tutorial and lecture material. Note that the *smallest* error is obtained when the measured response ( $y_0$ ) and concentration ( $x_0$ ) correspond to the centroid (mean of  $x$  and  $y$ ) of the data; the further the point from the centroid, the greater the error. This is illustrated in the graph below, which shows the results of three separate sets of calibration measurements obtained using the same standards, instrument, and method (the errors in each measurement have been exaggerated for the purpose of illustration.)

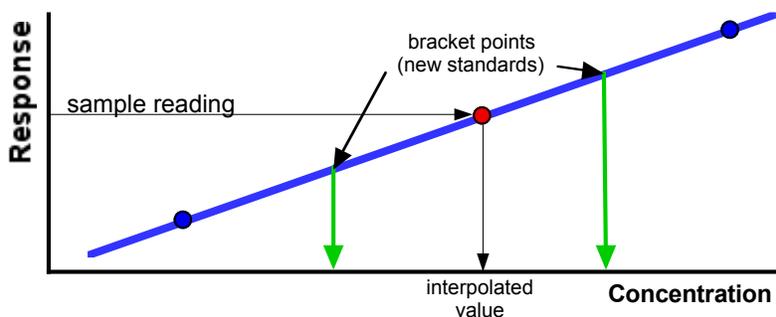


For an interpolated value, the standard error in that value is ( $m$  is the number of measurements for the sample value  $y_0$ ,  $n$  is the number of points in the calibration data, and other symbols are as defined above):

$$s_{x_0} = \frac{s_{y/x}}{b} \sqrt{\frac{1}{m} + \frac{1}{n} + \frac{(y_0 - \bar{y})^2}{b^2 \sum_i (x_i - \bar{x})^2}}$$

### 13.3 Bracketing the sample concentration:

One consequence of the increased uncertainty in interpolated values away from the centroid of the calibration data is that, in order to get meaningful results, it is often necessary to *bracket* your sample data points. This means that you obtain additional calibration points on either side of your experimental data, and perform regression analysis on this smaller sub-set of your calibration data. This is illustrated graphically below:



*Bracketing a sample point by adding new standards to the calibration curve*

Once you have identified target concentrations for additional standards, you can calculate the dilution factor required to obtain that value from one or more of your existing stock or standard solutions. You can then find the combination of transfer pipette and volumetric flask that most closely yield these values *without* using up large amounts of standard and solvent. For example, consider the following scenario:

Stock concentration,  $C_{stock} = 0.1005 \text{ M}$

Standards:  $C_1 = 0.001005 \text{ M}$ ,  $C_2 = 0.002010 \text{ M}$ ,  $C_3 = 0.005025 \text{ M}$ ,

$C_4 = 0.01005 \text{ M}$

Initial sample estimate:  $C = 0.00132 \text{ M}$

Since the sample is at the extreme end of the calibration range, we would want at least one extra standard midway between the first two at  $\sim 0.001507 \text{ M}$ :

$$\text{Dilution factor required using stock solution, } f = \frac{0.001507}{0.1005} = 0.01499 \approx 0.015$$

To get this, we could use a 15.00 mL pipette and a 1.000 L flask, but this would generate a lot of waste! We could also dilute 15.00 mL of the fourth standard (0.01005 M) to 100.0 mL, which is more reasonable:

$$C_{dilute} = C_4 \times \frac{V_{pipette}}{V_{flask}} = 0.01005 \times \frac{15.00}{100.0} = 0.001507_5 \text{ M}$$

### 13.4 Extrapolating a Value:

Normally in analytical chemistry we avoid extrapolating calibration curves for the simple reason that the uncertainties arising from the regression line are greatest at the extreme ends of the calibration range. The *relative error* will also be greatest where you have both large uncertainties and small values. Sometimes, however, specific techniques require the use of extrapolation in order to determine a value. One example of this is the method of standard additions. In its simplest form, this requires extrapolating a linear equation to the intercept on the  $x$ -axis,  $x_E$ , as shown on the next page.

The method works by first assuming that any interfering species in the sample will affect the signal due to the analyte by the same relative amount *regardless* of the analyte concentration. Suppose, for example, that the calibration curve for the analyte in the *absence* of any interferent was:

$$I = bC$$

If an interferent reduces the measured signal  $I$  by a fixed fraction  $f$ , then the calibration curve in the *presence* of the interferent will be:

$$I' = fbC$$

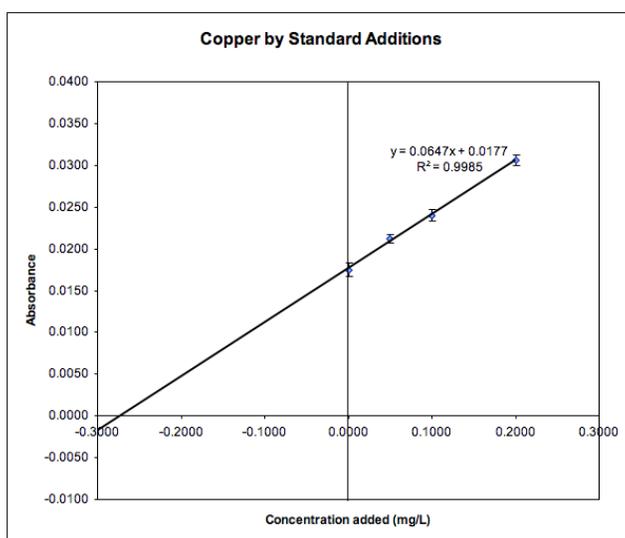
Note that when  $C = 0$ ,  $I = I' = 0$ . In standard additions, increasing amounts of a standard ( $C_s$ ,  $V_s$ ) are added to a fixed amount of sample ( $C_o$ ,  $V_o$ ). Assuming that each portion of sample with added standard is diluted to the same total volume ( $V_T$ ) then we can derive the equation:

$$I' = fbC = fb \left( \frac{C_o V_o}{V_T} + \frac{C_s V_s}{V_T} \right) = fbC_x + fbC_{add}$$

From this, we can see that a plot of  $I'$  versus  $C_{add}$  will have an intercept on the  $x$  axis ( $I' = 0$ ) equal to  $-C_x$ . The uncertainty, or standard error, of the intercept obtained in this manner can be calculated from the regression statistics as:

$$s_{x_E} = \frac{s_{y/x}}{b} \sqrt{\frac{1}{n} + \frac{\bar{y}^2}{b^2 \sum (x_i - \bar{x})^2}}$$

Once again,  $n$  is the number of points in the calibration curve,  $b$  is the slope, and the means refer to the mean  $x$  and  $y$  values for the calibration data.



Standard additions plot for the determination of copper by flame AAS