UNIVERSITY OF MALAYA

Master of Science

Instrumental Analytical Chemistry

Laboratory Manual

SQC7008
Advanced Laboratory Skill I
SQC7009
Advanced Laboratory Skill II

Department of Chemistry
Faculty of Science

DISCLAIMER

The instructions and synopsis stated in the lab manual is not exhaustive. In addition, the Program Coordinator, Course Coordinator, Lecturers and/or Laboratory Staff may alter or make changes to the statements in the lab manual from time to time during the process of the module. Any changes to the lab manual will be informed by the lecturer in charge of these modules.

Content

List of Experiment

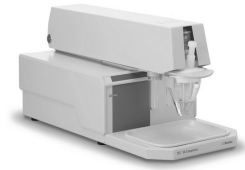
Introduction

SQC7008 and SQC7009 practical courses consist of a number of mini projects covering different aspects of the subject. The emphasis is to provide training in the analytical approach for the total analysis of samples of very different natures. It is hoped that students would gain valuable experience in a variety of both common and specialized techniques in following certain appropriate procedures in solving certain analytical problems of current interest.

You are advised to choose any two projects (from two different groups) and should plan to complete each project in about 20 hours. Once you have made the choice, you are required to look up for relevant literature, plan your experimental work and discuss the findings with the lecturer-in-charge of the class before proceeding any further.

Frequently, the solution to an analytical problem lies very much on one's analytical background and the ability to make good use of the immense literature in this field. This constitutes a very important element in the successful planning of your experiment. As general guide, each student should keep a note book for jotting down the literature search, planning of experimental works, observations and also the data acquired. The laboratory report must be submitted within 10 days after the completion of the mini projects.

Laboratory Report Writing Guide



Laboratory Report Writing Guide

(From: William, I., 2001.Environmental Chemistry: A modular approach, West Sussex, John Wiley & Soms, Ltd)

Introduction

As a student, you will be required to submit essays, laboratory and project reports to your lecturers for assessment. In future years, as a researcher, technician, teacher, academic, industrialist, civil servant, media correspondent, author, salesperson or politician, you may be required to write a range of scientific text targeted at a specific audience. This prospect may terrify you; many people regard writing as difficult, and something to be delayed or avoided. In fact, scientific writing is a skill, which, like tying your shoelaces or performing titration, can mastered with practice and perseverance. Like any other skill, scientific writing can be developed into something that will give you confidence, satisfaction and pleasure.

Laboratory reports are very important components of assessed work, and consequently, it is worth trying to produce good quality reports. As chemist, laboratory reports, are written for several reasons. One reason is to communicate the laboratory work to management. In such situations, management often bases company decisions on the results of the report. Another reason to write laboratory reports is to archive the work so that the work will not have to be done in the future. Laboratory reports are intended to demonstrate some or all of the following:

- you have performed and understood an experiment;
- you have some knowledge of the theoretical basis of the experiment;
- you can process/interpret the data obtained from an experiment;
- you can relate fundamental or derived laws to the outcome of the experiment;
- you can present these ideas/results in an appropriate context and can evaluate their significance.

Effective Scientific Writing

- Remember the purpose of your writing – communicate clearly, concisely and accurately.
- Consider your audience (tutor/lecturer) and the assessment criteria.
- Use appropriate format.
- Plan and arrange your ideas in a logical order.
- Treat what you write first as draft.
- Make sure your grammar, spelling and punctuation are correct.
- Ensure the first draft is clear enough.
- Re-read and edit your first draft as necessary.
- Proof-read the final draft, correcting any remaining mistake.

Grammar and Style

All the text in your report should be grammatically correct, properly punctuated and comprise complete sentences. The overwhelming majority of scientific reports are written using the impersonal Third Person / Past Simple Tense / Passive Voice form, acoiding, if possiblem the use of the personal pronounce (I, we, or you). The following examples illustrate what is intended:

Preferred "The samples were stored at 0 °C"

Not preferred "I stored the samples at 0 °C"

Presentation

Laboratory reports should be good to look at; a well-presented report will please the reader, give him/her confidence in the report and will aid assessment. A cover page will aid the presentation of your work, as well as providing important information to your assessor. The cover page should have:

- Course title and code;
- Number of experiment;
- Your report title;
- Your name and matric number;
- Name of your group members
- Date of submission;
- Name of Lecturer / Tutor.

Presentation (continued)

Laboratory reports should always use SI units. Unit is very important for all measurement. Without units much of our work as scientists would be meaningless. We need to express our thoughts clearly and units give meaning to the numbers we calculate. Knowing the units of measurement that correspond with a number can give you so much more information than a digit sitting there by itself. Units can:

- Help to show another person the exact amount you have;
- Assist in solving a mathematical problem, especially in chemistry, where you can follow the units to get to the answer;
- Show which measurement system the person is using (i.e. metric or standard).

Proper pagination of your reports will assist you to structure your work, as well as being good practice. It will also assist the reader / assessor to 'navigate' your report, thus making it easier to find relevant sub-sections, table, figures, etc. Pages containing preliminary information (e.g. cover page) are paginated in small Roman numerals (I, ii, iii, etc.), whereas pages of the main body of the report are given in Arabic numerals (1, 2, 3, etc).

Structure of the Laboratory Report

Basic structure for laboratory reports:

- Cover page (refer to section 4)
- Aims / Objectives of the Experiment
- Introduction
- Materials and Methods (Experimental)
- Results
- Discussion
- Conclusions
- References
- Appendices (if related)

Aims / Objectives of the Experiment

The aims or objectives of the experiment should clearly and briefly state the purpose of undertaking experiment. They usually include specific overall aims of the experiment. For example, in Experiment 4 that measures the oxygen content of water, the principal objective may be

- *To determine* the *dissolved oxygen* content of samples of *tap water* and *river water* using Winkler method.

You should always refer back to your aims in the Conclusions section of your report and comment upon whether they have been achieved satisfactory.

Introduction of the report

The introduction should establish the context of the experiment, and explain the rationale for undertaking it (i.e. why is it worth doing at all). Here, you should provide some background information on the problem under investigation, such as the source of the pollutant under investigation and any potential health/environmental effects. This section can also involve a description of the theory relating to the experiment and the experimental technique(s) to be used. It should leave the reader with the feeling that the report has a general relevance and that to read on would be worthwhile.

Materials and methods

This section should contain a concise but adequate description of all of your experimental materials and procedures so that your results could be verified independently. Materials, too should be as fully described as is necessary for replication. The details of the apparatus / instrument (e.g. UV-Vis Spectrophotometer; GC-FID, AAS, etc) used should be included at this section. There is also no need to repeat routine instructions for using apparatus or equipment where they are well-known or available in manufacturers' instruction. Fig 1 shows the example of the description for chemicals and instruments.

Any form of sampling procedures must be very fully described – both the sampling techniques and the sampling strategy. Sampling usually undertaken to obtain some estimate relating to a population. Similarly, locations and study areas

2.1. Chemicals

Parabens (esters of 4-hydroxybenzoic acid, MeP, EtP, PrP, BuP and BzP), phenol and nitrobenzene (NB) were obtained from Fluka. *Tert*-butanol (*t*-BuOH) was obtained from Sigma–Aldrich. All solvents (Merck) were of the HPLC grade. Individual parabens stock solutions were dissolved in boiled ultrapure deionized water (Elga, USA). A mixture of BSTFA (*N,O*-bis(trimethylsilyl)trifluoroacetamide) and TMSCl (trimethylchlorosilane) in a ratio of 99:1 was obtained from Supelco (USA). Sodium phosphate monobasic and Sodium phosphate dibasic were purchased from Sigma and Riedel-de-Haén, respectively.

2.5. Instrumental

All HPLC analyses were performed using Shimadzu HPLC system consisted of a LC-20AT pump, a SPD-M20A diode array detector, a SIL-20AHT auto sampler, a CTO-20AC column oven and a CBM-20A communication bus module (Shimadzu, Japan). A reversed-phase Chromolith RP-18 monolithic column (100 mm × 4.6 mm; Merck, Germany) was used for separation.

Analysis of degradation by-products was carried out using a Hewlett–Packard Model 6890 GC, with a HP-5 (5% phenylmethylpolysiloxane) column. The detail of the setting and the GC temperature program was given in previous study (Tay et al., 2009).

Fig 1 Example of writing the description for chemicals and instruments

Reporting Results

Clearly, the Results are an exceptionally important part of your report and great care should be taken in their presentation. Over the years, a number of conventions have developed in the reporting of results. It is important to open your Results section with appropriate text rather than by just presenting tables of data. A table must follow, and never precede, the frist reference to it in the text. You should not leave it to the reader to interpret tables – that is your job. An acceptable format is of the type, 'The data presented in Table 1 show that ……'. Indeed, the reader should be able to appreciate the significance of the result without reference to any talbe of data; the data are evidence to support your statements. While tables are used to present the data, figures can be helpful in interpreting them.

Tables

Tables are the main vehicles for conveying data to the reader. A table can be considered as a complete entity, in a sense, should be able to exist separately in the text. A well-constructed table does not need a lengthy explanation on how it is to be interpreted but should be self-explanatory and be characterized by its simplicity and unity. The caption (on top of the table) is clearly important if the table is to stand as a separate entity. Table 3 is a well laid out and clear example.

Figures / Graphs

Laboratories exercises will oftern involve the production of graphs from the data collected. A graph can provide much more information than a set of data. It gives a visual representation of trends and relationships, and permits the prediction of what happens between the known points. Graphs are commonly labeled as Figure in lab reports. As tables, appropriate captions (or titles) should be added at the bottom of the graph (refer to Fig. 3). Well-drawn graphs can greatly enhance the effectiveness of display and interpretation of the results presented in a report.

Table 3. Characterization of the leachate collected from the Gramacho Metropolitan Landfill used in this work (n = 4 samples).

Parameters	*Average value*	*Standard deviation*
pH	8.3	0.3
Total alkalinity (mg $CaCO_3$ L^{-1})	8857	1480
Carbonate alkalinity (mg $CaCO_3$ L^{-1})	450	490
Bicarbonate alkalinity (mg $CaCO_3$ L^{-1})	8374	1917
Total ammonia nitrogen (mg [N-NH_3] L^{-1})	1998	387
Chloride (mg L^{-1})	3196	862
Dissolved Reactive Phosphorus (mg L^{-1})	7.5	1.3
Total Solids (mg L^{-1})	9390	2087
Total Suspended Solids (mg L^{-1})	53	31
DOC – dissolved organic carbon (mg L^{-1})	935	71
COD chemical oxygen demand (mg L^{-1})	3332	523
BOD - biochemical oxygen demand (mg L^{-1})	141	45

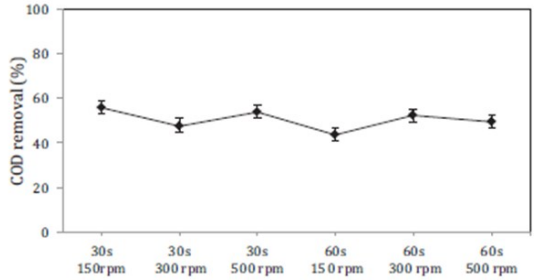


Fig. 3. Removal of COD for various mixing times and stirring speeds (experimental conditions: pH = 4.0, $FeCl_3$ dosage = 1400 mg L^{-1} and temperature = 23°C).

Discussion

The Discussion should draw all the threads of the report together and is, arguably, the most important part of the report. The discussion offers the widest scope for individual freedom of expression, and may include items such as the following:

- A comparison of the results with those obtained or published elsewhere;
- A discussion of the significance of the data in an appropriate context;
- Comments on the value of the results in a wider scientific, environmental or even commercial context.
- A discussion of the possible limitations of the methods;
- Comments upon the precision, reproducibility or repeatability) of the results, as well as on the accuracy, if known.
- A discussion of effectiveness and limitations of the experiment and any statistical treatment of the data.

Attention should be drawn to any fault/problems with the chemicals or equipment used and to any deficiency in the assumptions upon which the experiment is based. Modifications and improvements should be included if appropriate.

Conclusions

The Conclusion section should summarize the main findings of the experiment. It is not a summary of your work programme or a description of the research carried out. It is often helpful to use 'bullt points', each no more than two or there lines, to summarize your results. This enables you, lecturer and tutor to see, at glance, whether you have addressed all of the important areas and helps you to check that you have covered everything that you wanted to and listed in the objectives.

References

Citing references

References may be cited in the text in a number of ways, depending upon your style of writing or the context of your reference. However, there are convention that should be followed, as shown below – note the use of brackets.

Natural levels of carbon monoxide are low, typically in the range 20 – 200 ppb (Grimes and Clement, 1993).

Kinnear (1998) describes a system for sampling PM10 on an hourly basis, while Hegarty et al. (2001) describe a system for the continuous sampling of PM10. [Note: "Hegarty, Scanlon and Chan (2001) is written as Hegarty et al. (2011)]

If reference has two authors or less, the family name of all author(s) should be mentioned in the reports. If a reference has more than two authors, only the first is mentioned with "et al." "et al. translates as "and others".

You may want to cite an official or company report, or government paper, where there is no specified author or the authorship belongs to a committee. In such cases, you normally cite the body responsible for publishing the paper or report. Thus, in the text, the body responsible for publishing the paper is cited with the year of publication, e.g. (EvironTech Ltd, 2000).

The Reference Section

The Reference section must include details of all references ***that have been cited in text***. It does not include peripheral reading. The details of each reference include the following: name(s) of the author(s) (surname first, with a comma), the year of publication, and the title of the publication. In the case of books and reports, the name of the publisher and place of publication is also given, There is more than one way of presenting this information; the following example illustrate the use of upper and lower case letters, italics, punctuation marks and general layout.

Books

Example: Roberts, M.B.V. (1984). Biology: A Functional Approach (3rd Edn). Nelson Publishers, London.

Book Chapters

Example: XYZ, F.M. (Year Published). Title of chapter In F.M. XYZ Editor (Ed.), Title of book/anthology (pp. Pages). Publisher City, State: Publisher.

Article in Journals

Example: XYZ, F. M., & ABC, F. M. (Year Published). Article title. Journal Name, Volume (Issue), Pages.

Websites

Example: Satalkar, B. (2010, July 15). Water aerobics. Retrieved from http:// www.buzzle.com

PLAGIARISM

Plagiarism is the representation of another person's published or unpublished work or ideas as your own by using an extensive unacknowledged quotation. In academia, plagiarism carries heavy penalties; your mark for any assessd work may be significantly reduced and you may be open to accusations of academic misconduct. However, this does not mean that all of your work must be completely original; expressing views that are influenced by other authors is a consequence of shared knowledge and reflection of wide reading. In order to avoid accusations of plagiarism, you should clearly reference sources by using the conventions outlined above.

Marking Rubric

Component 1: Lab Performance (Total 20%)

Pre-entering lab (5%)

Score	Criteria
0	• No preparation of experimental procedure. • No proper attire (shoes, safety glasses, lab coat).
1-3	• Procedures are too brief, lack of details and confusing. • Incomplete safety attire.
4-5	• Presents easy to follow steps in lab experimental, logical and adequately detailed. • Complete safety attire checked.

Skill and Techniques (15%)

Score	Criteria
0	• No skill is demonstrated.
1-5	• Wrong glassware used, wrong technique, spillage and wasting of chemicals.
6-10	• Right glassware used, incorrect or lack of lab technique.
11-15	• Presents correct lab skill, clean and tidy.

Component 2: Report (Total 60%)

Section	Rubric	
Title (5%)	0-1	No title, or Too brief (e.g. "Lab report"; "Mercury in fish"; Ascobic acid in fruits", etc).
	2-3	Too long, or Does not identify the complete subject of study
	4-5	Identify the complete subject of study and encapsulates the purpose of the report/study.
Objective (10%)	0	Section missing completely.
	1 - 5	Be too vague, ambitious or broad in scope; Just repeat each other in different terms; Just be a list of things related to the topic; Contradict with methods; Does not identify subject of study.
	6 - 10	Concise and brief; Be interrelated and describes how to achieve that objective; Clearly identify the subject of study; Related to the experiment that has been done.
Introduction (15%)	0	Section missing completely.
	1 - 5	Very short with no attempt to include any references to relevant work.
	6 - 10	Has include everything that relevant to the work with some evidence of extra reading, but the structure isn't very clear and appears disjointed, or Well written with no evidence of any extra reading.
	11 - 15	Clearly written, well structured, with evidence of extra reading; Clear outline of study's hypotheses; Does show something novel in it as compared to the supplied handout/laboratory manual; Does include the rationale for performing the experiment.
Experimental (15%)	0	Section missing completely
	1 - 5	One or more subsections (e.g. chemicals or instrumentation) are missing; Confusing statement; Parts have been included under the wrong sub-section.
	6 - 10	Somewhat confused; Parts are missing within subsections; Parts have been included under the wrong subsection.
	11 - 13	Good structure, but some relevant information has been omitted.
	14 - 15	Contains all of the relevant information about the method used; clearly and systematically described in such a way that a reader could replicate the study from the description.

Component 2: Report (Total 60%) (continued..)

Section	Rubric	
Results (20%)	0 - 5	Graphs or tables are included without caption and any written explanation; Has some writing without tables and graphs; Very poor presentation of the collected data.
	6 – 10	Has included the raw data in tables; Poor presentation of data (e.g. no graphs; wrong graphs; irrelevant graphs, no label and caption); Inaccurate explanations
	11 – 15	Has presented the data in a logical format (e.g. graphs, tables); Show some understanding with explanation, but some relevant information has been omitted; Graphs and tables are not label accordingly/correctly; Standard deviations or standard errors missing from the presented data (if related)
	16 – 20	Logical sequence; Clear presentation with relevant and clear explanation; Figures and tables are well labeled
Discussion (20%)	0 - 5	No attempt to relate results to relevant theoretical and empirical research; Does not understand of what the study was about.
	6 – 10	Poor structure, wrong order, shows little understanding of the experiment.
	11 - 15	Poor structure, but contains essential elements, or Good structure with some missing elements
	16 – 20	Well organized and clearly written; Clearly summarize the obtained results; Does show attempt to relate the findings to previous research; Does show ability to evaluate the weakness and limitations of the study; Does include sensible suggestion for possible improvement.
Conclusions (10%)	0	Section missing completely
	1 – 5	Conclusion is drawn but not supported by experimental evidence; No sensible conclusion is drawn; No clear evidence of a thorough understanding of the experiment and/or theory behind the experiment.
	6 – 10	Conclusion is drawn and supported by experimental evidence; Sensible conclusion is drawn; Shows clear evidence of a thorough understanding of the experiment and/or theory behind the experiment.

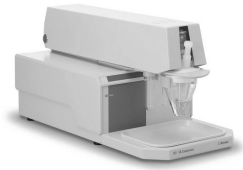
Component 2: Report (Total 60%) (continued..)

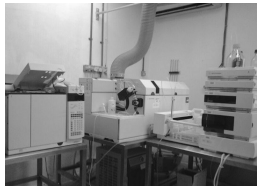
Section	Rubric	
References (5%)	0	Reference not included in the report
	1 - 3	Incomplete references to the books or any other sources used in report.
	4 - 5	References in the text and in the reference list conform in all respects to the formatting convention (e.g. APA format); Complete references to the books or any other sources used in report; References in text are matched with references in reference list (e.g. no missing references)

Component 3: Assessment of understanding on the conducted experiments (20%)

Score	Criteria
0	• Unable to answer any questions.
1-5	• Very little attempt to answer question correctly.
6-10	• Most answers are incorrect, and some are irrelevant to the question type.
11-15	• Some answers maybe very short or incomplete.
16-20	• Questions are answered to the best of abilities and answers match the question types.

Synopsis





Chromatographic Analysis - GC

Experiment 1

PREPARATION OF FATTY ACID METHYL ESTERS (FAMEs) AND DETERMINATION OF FATTY ACID PROFILE OF OILS BY

Synopsis:

Information about fatty acid profile on food is important for nutrition labeling, which involves the measurement of not only total fat but also saturated, unsaturated, and monounsaturated fat. Gas chromatography is an ideal instrument to determine (qualitatively and quantitatively) fatty acid profile or fatty acid composition of a food product. This usually involves extracting the lipids and analyzing them using capillary gas chromatography. Before such analysis, triacylglycerols and phospholipids are saponified and the fatty acids liberated are esterified to form fatty acid methyl esters (FAMEs) so that the volatility is increased.

Two methods of sample preparation for FAMEs determination will be used in this experiment: (1) sodium methoxide method, and (2) boron trifluoride (BF_3) method. In the sodium methoxide method, sodium methoxide is used as a catalyst to interesterify fatty acid. This method is applicable to saturated and unsaturated fatty acids containing from 4 to 24 carbon atoms. In the BF_3 method, lipids are saponified, and fatty acids are liberated and esterified in the presence of a BF3 catalyst for further analysis. This method is applicable to common animal and vegetable oils and fats, and fatty acids. Lipids that cannot be saponified are not derivatized and, if present in large amount, may interfere with subsequent analysis. This method is not suitable for preparation of methyl esters of fatty acids containing large amounts of epoxy, hydroperoxy, aldehyde, ketone, cyclopropyl, and cyclopentyl groups, and conjugated polyunsaturated and acetylenic compounds because of partial or complete destruction of these groups.

Objective

- Utilize two methods to prepare methyl esters from fatty acids in food oils.
- Determine the fatty acid profile and their concentration in the oils by gas chromatography-flame ionization detector (GC-FID).

References

Min DB, Ellefson WC (2010) Fat analysis Ch. 8. In: Food analysis, 4th edn. Springer, New York.

O'Keefe SF, Pike OA (2010) Fat characterization, Ch. 14. In: Nielsen SS (ed) Food analysis, 4th edn. Springer, New York.

Chromatographic Analysis - IC

Experiment 2

DETERMINATION OF ANIONS IN SOIL SAMPLES USING ION CHROMATOGRAPHY (IC)

Synopsis:

Soil is the natural medium in which the roots of most plants grow. From soil, the plant adsorbs water and solutes necessary for its continued well-being. If soil is fertile, it contains in a readily available form of all the chemical elements essential for plant growth. Nitrogen, phosphorus and sulfur are three important nutrients by plants. They are taken up by plant roots from soil solution as SO_4^{2-} , NO_3^- and H_2PO^{4-}. The analysis of these anions is important in order to investigate the quality of soil.

Ion chromatography (IC), a liquid chromatographic methods based upon ion exchange reactions, has been used to determine a variety of ions in soil extracts. Ion chromatography is a rapid, sensitive, and precise method which can simultaneously detect all the target analyte.

The determination of SO_4^{2-}, NO_3^- and H_2PO^{4-} in soils can be detected with ion chromatography (IC) which requires appropriate sample preparation prior to analysis. The extractant type during sample preparation of soil might be different depends on anions due to the affinity of some of anions commonly present in soil is of the order: $NO_3^- < Cl^- < SO_4^{2-} < PO_4^{3-}$.

Objective

- To determine selected anions in soils using ion chromatography.
- To compare different type of extractants during sample preparation of soils.

References

Dick, W. A., & Tabatabai, M. A. (1979). Ion chromatographic determination of sulfate and nitrate in soils. Soil Science Society of America Journal, 43(5), 899-904.

Jackson, P. E. (2000). Ion Chromatography in Environmental Analysis. Encyclopedia of Analytical Chemistry (Meyers, R. A. (Ed.)), pp.2779-2801.

Mussa, S. A. B., Elferjani, H. S., Haroun, F. A., & Abdelnabi, F. F. (2009). Determination of available nitrate, phosphate and sulfate in soil samples. International Journal of PharmTech Research, 1(3), 598-604.

Nieto, K. F., & Frankenberger, W. T. (1985). Single column ion chromatography: I. Analysis of inorganic anions in soils. Soil Science Society of America Journal, 49 (3), 587-592.

Stanišić, S. M., Ignjatović, L. M., Stević, M. C., Đorđević, A. R. (2011). A comparison of sample extraction procedures for the determination of inorganic anions in

Chromatographic Analysis - IC

Experiment 3

DETERMINATION OF OXALATE AND SOME INORGANIC ANIONS IN TEA FUSIONS BY ION CHROMATOGRAPHY

Synopsis:
This experiment aims to quantify oxalate ion and other inorganic anions in tea fusions by using ion chromatography (IC). Oxalate can be found in food products such as sorrel, chocolate, cacao and tea. After water, tea is the second most popular beverage, but it contains high levels of oxalic acid. In the human body, oxalic acid is formed by the metabolism of glycine and ascorbic acid. The role of oxalic acid in the human body is very significant, because its compounds are responsible for the stability of biological membranes. However, insoluble calcium and magnesium oxalates may be accumulated in the body in the form of kidney stones. Therefore, it is necessary to control the concentration of oxalic acid in food and the human body. Nowadays, oxalate ions can be identified in complex objects using titration, colorimetry, capillary electrophoresis (CE), chemiluminescence, high performance liquid chromatography (HPLC), gas (GC) and IC. However, these methods have disadvantages. CE has low reproducibility due to migration times of ions. GC involves difficult sample preparation. Since oxalic acid is a strong organic acid (pKa1 = 1.27; pKa2= 3.80), the most effective method for the determination of oxalate ions is IC with conductometric detection. In this work, an approach for the simultaneous determination of oxalate, chloride, nitrate, phosphate, sulphate ions should be proposed and applied to the analysis of anions in tea samples of different brands and manufacturers.

Objective

- To to quantify oxalate ion and other inorganic anions in tea fusions by using ion chromatography (IC)

References

Yusenko, E., Polyntseva, E., Lyzhova, A., Kalyakina, O. (2013) Determination of oxalate and some inorganic anions in green and black tea. Proceedings of the Latvian academy of Sciences, Section B, Volume 67, No. 4/5 (685/686), pp. 429-432.

Chromatographic Analysis - HPLC

Experiment 4

DETERMINATION OF PARABENS IN COSMETIC CREAMS BY HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC)

Synopsis:

Preservation of cosmetic products is necessary in order to prevent alteration and degradation of the formulation through microbial contamination, and to protect the consumers. Parabens are commonly used as antimicrobial reagents in cosmetic and pharmaceutical products. The quantitative analysis of each individual additive is an important issue of quality assurance in commercial products.

The ester derivatives of p-hydroxybenzoic acid (parabens) are antimicrobial mostly used in the cosmetic industry. Published methods for the analysis of parabens in cosmetic products are based on gas chromatography (GC) and liquid chromatography (LC). Among them, GC methods are most often employed for complex matrices. However, they often require derivatization prior to injection and thus are more time-consuming than LC. In LC, aqueous samples are injected onto the column without prior partition. Therefore, the solid and emulsified products which are often encountered in the analysis of cosmetics must first be converted into a liquid form before LC analysis.

Objective

- Develop a HPLC separation method for the determination of parabens in cosmetic creams.
- Determine the concentration of parabens in cosmetic creams.

References

Min DB, Ellefson WC (2010) Fat analysis Ch. 8. In: Food analysis, 4th edn. Springer, New York.

O'Keefe SF, Pike OA (2010) Fat characterization, Ch. 14. In: Nielsen SS (ed) Food analysis, 4th edn. Springer, New York.

Qian M, Peterson DG, Reineccius GA (2010) Gas chromatography. Ch. 19. In: Nielsen SS (ed) Food analysis, 4th edn. Springer, New York.

Qian MC (2015) Gas chromatography. Ch. 19. In: Nielsen SS (ed) Food analysis Laboratory Manual, 2th edn. Springer, New York.

Elemental Analysis - UV-Vis Spectroscopy

Experiment 5

IRON DETERMINATION IN MEAT USING UV-VIS SPECTROSCOPIC METHOD

Synopsis:

Chromogens are chemicals that react with compounds of interest and form colored products that can be quantified using spectroscopy. Several chromogens that selectively react with minerals are available. In this lab, ferrozine is used to measure ferrous iron in an ashed food sample. The relationship between the absorbance of the chromogen-mineral complex is described by Beer's Law; in this procedure, a standard curve is generated with a stock iron solution to quantify the mineral in beef samples.

In this experiment, meat samples are first ashed to dissociate the iron bound to proteins, and the ash residue is solubilized in dilute HCl. The acid is necessary to keep the mineral in solution. Ferrozine complexes only with ferrous iron and not with ferric iron. Prior to the reaction with ferrozine, the solubilized ash is first treated with ascorbic acid to reduce iron to the ferrous form. This step is necessary with ashed samples, as this procedure would be expected to oxidize all the iron present in the meat. However, when other treatments are used to liberate iron, for example, trichloroacetic acid precipitation, a comparison of samples treated with ascorbic acid and untreated samples could be done to determine the ratio of ferrous to ferric iron in foods.

Objective

- Determination of Iron in meat samples by using spectrophotometric method

References

Ward, R.E. and Carpenter, C.E., 2010. Traditional methods for mineral analysis. Ch. 12, in Food Analysis, 4th ed. S.S. Nielsen (Ed.), Springer, New York.

Carpenter, C., Ward, R., 2015. Iron determination in meat using ferrozine assay, in Food Analysis Laboratory Manual, 2nd ed. S.S. Nielsen (Ed.), Springer, New York.

Molecular Spectroscopy- UV-Vis Spectroscopy

Experiment 6

MEASUREMENT OF CAFFEINE IN COFFEE BEANS WITH UV/VIS SPECTROMETER

Synopsis:

Caffeine is found in various kinds of foods and drinks that we consume in daily life. It causes various physiological effects such as relaxation of bronchial muscle, stimulation of the central nervous system, gastric acid secretion and dieresis. And their concentration in vivo is a key mark for various disorders including heart disease, carcinogenesis, kidney malfunction and asthma. On the other hand, chemical analysis of caffeine in coffee beans is also used as an additional tool for evaluating coffee quality. Higher caffeine contents associated with highest quality samples compared to other Arabic samples have been reported. Therefore, establishing a rapid and cheap analytical method for the determination of caffeine in coffee beans has an interest for a wide range of physiological effects on the human body and quality controls.

Several chemical and physical methods have been developed for the determination of caffeine in coffee and other beverages. The most widely used methods for the determination of caffeine in beverages include various analytical techniques such as derivative spectrophotometer, HPLC, Fourier Transform infrared, NIR reflectance spectrometry, Raman spectroscopy and capillary electrophoresis, which have been reported. Although Spectrophotometer is a fast and simple method it is not possible to determine caffeine directly in coffee beans by conventional UV absorption measurement due to the spectral overlap. On the other hand, the derivative spectrophotometer is relatively easy; however, it is not reliable for the determination of small concentration of caffeine in samples. In this experiment, you are required to develop a method to quantify caffeine in coffee bean/powder using UV/Vis spectrophotometer, which is available in most laboratories.

Objective

- To develop a method to quantify caffeine in coffee bean/powder using UV/Vis spectrophotometer.
- To quantify caffeine in different coffee bean/powder samples.

References

Belay, A.., Ture, K., Redi, M., Asfaw, A. (2008) Measurement of caffeine in coffee beans with UV/vis spectrometer. Food Chemistry 108, 310–315.

Elemental Analysis - AAS

Experiment 7

DETERMINATION OF MINERAL CONTENT IN FOOD PRODUCTS/ SUPPLEMENTS BY ATOMIC ABSORPTION SPECTROSCOPY

Synopsis:
Dietary minerals are inorganic nutrients that essential to maintain human health, as they can be regarded as chemical elements that support human biochemical processes. Adequate intake of these elements ensures nutritional quality of one's diet. In order to protect consumer interests, the Food Regulations 1985 mandates nutrition labelling for wide variety of foods sold in Malaysia which means quantification of mineral content of the products is mandatory. The task is common accomplished by the technique of atomic absorption spectrometry which requires appropriate sample treatments prior to analysis.

Dry ashing and wet ashing are two primary sample treatment procedures in the determination of mineral content of food samples. In dry ashing, the organic matrix of the sample is removed by incineration in present of oxygen at a high temperature, whereas, in wet ashing, the sample is digested with a mixture of concentrated strong acid. These methods can be accelerated using microwave technologies.

Objective
- To determine selected mineral contents of food products using flame atomic absorption spectroscopy.
- To compare the results obtained by different means of sample preparation.

References
USEPA (1996) Method 3052, U.S. Environmental Protection Agency, Washington.

Park YW and Bell LN (2004) Determination of moisture and ash content of foods. In: Nollet LML (ed) Handbook of Food Analysis: Physical characterization and nutrient analysis, Marcel Dekker Inc, New York.

Nielsen SS (2015) Sodium and Potassium Determinations by Atomic Absorption Spectroscopy and Inductively Coupled Plasma-Atomic Emission Spectroscopy. In: Nielsen SS (ed) Food analysis Laboratory Manual, 2th edn. Springer, New York.

Elemental Analysis - CVAAS

Experiment 8

DETERMINATION OF TOTAL MERCURY IN COSMETIC PRODUCTS BY COLD VAPOUR ATOMIC ABSORPTION SPECTROMETRY (CVAAS)

Synopsis:
Cosmetic can be regarded a global practice where skin-lightening constituted an important element in cosmeceutical industry. Among the ingredients, mercury is a well-documented melanotoxin added to some lightening products. However, chronic mercury exposure can cause health problems include dermatologic, renal, and neurologic issues. Thus, the mercury content in such products must be verified against possible adulteration as they are subjected restrictive regulations such as the Malaysian Control of Drugs and Cosmetics Regulations 1984.

Depends on its formulation, the matrix of a cosmetic product is not simple in most cases. It requires appropriate sample treatments priori subjected to the mercury determination technique. In many laboratories, the sample dissolution is generally accomplished by the uses of strong oxidizing mixture, high temperature and high pressure before being introduced to cold vapour atomic absorption spectrometry. Subjected to the nature of the sample solution, the calibration involves either external calibration or standard addition approach.

Objective
- To determine mercury level in cosmetic products by cold vapour atomic absorption spectrometry.
- To demonstrate the performance characteristics of the analytical method are fit for purpose.

References
USEPA (1993) Method 254.5, U.S. Environmental Protection Agency, Cincinnati.

Peregrino CP et al. (2011) Int J Environ Res Public Health 8: 2516.

Ng SY et al. (2015) Int. J. Mass Spectrom. 389: 59.

Elemental Analysis - ICP

Experiment 9

ELEMENTAL FINGERPRINTING OF SOILS USING INDUCTIVELY COUPLED PLASMA MASS SPECTROMETRY

Synopsis:
Soils are composed of inorganic compounds and organic matters where the proportions between both components can vary widely. The chemical variations of soils originate mainly from variability in the ratio of both components in soils and the chemical variability in the inorganic fraction. In term of elemental composition, thus, the distribution pattern varies from soil to soil where the inorganic faction usually associated with greater variations than the organics. Part of the elemental variation is inherited from the parent materials which the soils developed and results from natural processes over time. To certain extent, additional deviations in the elemental composition of soil are attributed by anthropogenic impacts. Such elemental distribution pattern is an important characteristic of soils which could be used for fingerprinting and traceability purposes.

The techniques used in elemental fingerprinting are mostly those with multiple-element detection capability; where inductively coupled plasma mass spectrometry has been acknowledged as a premier technique for simultaneous isotopic determination of a wide range of elements. With the support of chemometric analysis, the underlying elemental patterns that inherent with soil samples can be revealed, and this is essentially helpful when the number of the characteristic elements increases.

Objective
- To determine multi-elemental concentrations of different soil samples using inductively coupled plasma mass spectrometry.
- To evaluate the elemental distribution patterns using chemometric tools.

.

References
USEPA (2007), Method 3051A, U.S. Environmental Protection Agency, Washington.

Low K.H. Food Anal. Methods (2011) 4:276.

Reidy L et al. (2013). Forensic Sci. Int. 233(1–3): 37.

Chromatographic Analysis - GC FPD

Experiment 10

DETERMINATION OF ORGANOPHOSPHORUS PESTICIDE RESIDUES IN FRESH FRUITS AND VEGETABLES BY GAS CHROMATOGRAPHY

Synopsis:
Pesticides have numerous beneficial effects but also have a risk. Pesticides are toxic, contain any substance, or mixture of substances of chemical or biological ingredients and it was designed to kill, reduce or repel insects, weeds, rodents, fungi or other organisms that can threaten public health and the economy. The pesticides used by the farmers is to control their fruits and vegetables from disease and pest and at the same times it will help them to get high yield of the plants. Organophosphorus pesticides (OPPs), widely known as persistent organic pollutants, are the most popular contaminants in agriculture products in developing countries. For this reason, many countries have established monitoring programs and legal regulations to control the use of pesticides on edible crops. Pesticides residues must comply with the Maximum Residue Levels (MRLs) established by national and international regulations.

Most of the analytical method used in the determination of OPPs are based on chromatographic technique such as using gas chromatography (GC) for volatile and thermally stable analyte or using high-performance liquid chromatography (HPLC) for involatile and low thermal stability analyte.

Objective

- Develop a GC-FPD separation method for the determination of OPPs in fresh fruit and vegetable samples.
- Determine the concentration of OPPs in fresh fruit and vegetable samples.

.

References

Sapahin, H. A., Makahleh, A., & Saad, B. (2014). Arab. J. Chem.

Amiri, A., Tayebee, R., Abdar, A., & Narenji Sani, F. (2019). J. Chromatogr. A. 1597: 39-45.