

Most laboratory and field analysis involves the use of pre-existing methods, without any need for modifications. However, there will be occasions in most technicians' work where a new analyte or sample is to be analysed, or a new method is to be investigated.

When this occurs, some pre-planning and possibly literature research will be required before any practical work is done. In the most extreme case, a technician may have to develop a new method almost from the ground up. However, there is very few analyses that haven't been done before, so in most cases, the research literature will contain work done on the same task or one very similar. More often, what will be needed is to **choose** between alternative procedures (perhaps a proposed new and the existing one) or to **adapt** an existing one.

Before we go any further, the meaning of a few terms should be clarified: *method*, *procedure* and *technique*. For our purposes, the first two terms mean the same thing: a method or procedure is the complete set of steps required to analyse a sample, including sample and standard preparation, while a technique is the specific analytical process/instrument, such as titration or AAS.

The process of developing any analytical method can be summarised into the following steps (see also Figure 1.1):

1. **Research into existing procedures** – there's no point in re-inventing the wheel so seeing what is already there will save a lot of time
2. **Define the task** – identifying what it is about the sample and the laboratory that is important in determining the choice of procedure,
3. **Choose a procedure** – based on knowledge of the advantages and disadvantages of the various methods as they relate to the task (see point 2),
4. **Adapt the existing procedure to suit the sample** – it will be rare that someone else's procedure will be a perfect fit for your situation
5. **Check the procedure** – running through the method with the sample, and also by some checking procedure to test its accuracy, and finally
6. **Correct problems** - small or large variations for problems discovered during the checking.

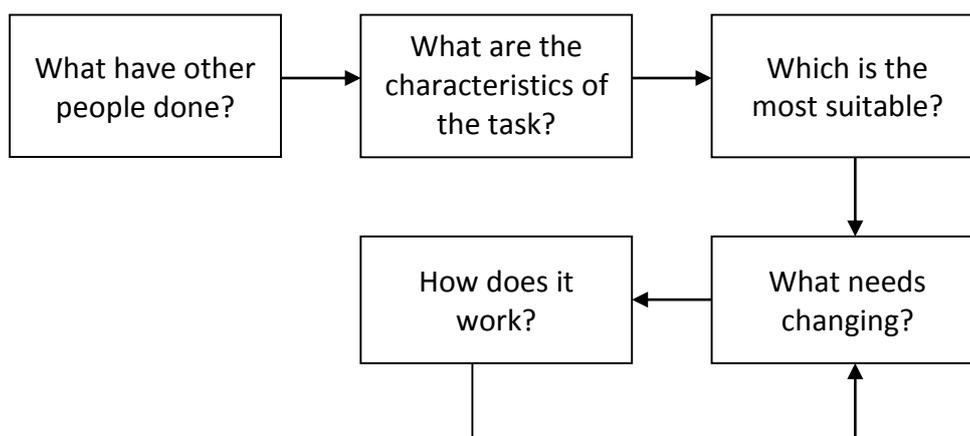


FIGURE 1.1 The process for developing a new procedure

1. Searching the analytical literature

Most analyses have been done before, or at the very worst, something closely resembling what you want to do. The hard part is finding the information in the vast quantity of published work. Where do you start?

Firstly, the two types of information you will be after are *how to do the analysis* and *how much analyte you can expect to find*. Sometimes, the two are in the same publication, but more often they won't be.

Basically, there are three general sources of useful information (hardcopy or Internet):

- *background information*, such as texts, providing theory on techniques, and data books, providing typical analyte concentrations,
- *collections of proven methods*, ranging from the general (analyte in anything) to the specific (analyte in specified sample), with supposedly few if any "bugs", and a clearly written procedure
- *research publications*, documenting original research into techniques and methods, not necessarily bug-free or clearly written.

Some of the more useful sources available on campus are given below in Table 1.1. Internet searches for information will be discussed below.

TABLE 1.1 Literature sources of information

Book	Information
The Composition of Foods (McCance & Widdowson)	comprehensive list of food products and their typical composition
Vogel's Textbook of Quantitative Analysis ("Blue Vogel")	reference collection of theory and practice for general purposes analyses, some specific analyses are discussed
Australian Standards	all types of testing and requirements, including chemical analysis; these are the tests that labs must follow for special registration
Pearson's Chemical Analysis Of Foods	analysis of foods, with background information and typical compositions
Standard Methods Of Analysis	physical and chemical methods of analysis for most materials
Official Methods Of Analysis	primarily chemical methods across a range of materials, primarily foods, pesticides, pharmaceuticals etc, not for metals or water
Encyclopaedia Of Industrial Chemical Analysis	provides wide range of methods and information about each
Standard Methods for Analysis of Waste and Wastewater (APHA)	as the name implies, this concentrates on environmental analysis of water samples; most environmental laboratories and standards refer to this
Rump & Krist	wide-ranging coverage of various environmental tests
green soil book	comprehensive detail of most common soil test methods

Instrument manufacturers

Most instruments purchased these days come with a useful manual, which provides some typical analytical procedures, covering a range of analytes and samples. The Varian AAS manuals are particular good in this regard.

Research journals

The world's store of knowledge regarding chemical analysis has been developed by research facilities: university, government or private. The outlet for this information is the huge number of journals (specialist magazines) that publish original research. The amount of scientific research published in journals is truly staggering. Much of it has no direct use, and may never have, but pure academic research is still essential, because many extremely important scientific breakthroughs came after scientists studying something just to see how and why it is as it is. Someone else then used that knowledge to develop something "useful".

In the analytical chemistry field, two of the most important are *Analytical Chemistry* and *Journal of AOAC International*. The former is the premier journal, but is aimed at pure research, and is therefore often very obscure and difficult to understand. The latter is the journal associated with the *Official Methods Of Analysis* collection, and is more applied.

Finding what you need in the sea of publications would be near impossible without some means of summarising what is available. This is what is known as **Abstracts**: indexes of all the journal articles published in a particular time period in a particular scientific area, eg *Chemical Abstracts* and *Analytical Abstracts*.

The Internet

You might think that in three billion Internet pages, there must be some really useful information on your task. It won't necessarily be so. At best, you might find some information about expected concentrations, but the chances of finding a fully detailed method are quite remote.

The most useful source of information on analytical procedures on the WWW will probably come from the major instrument manufacturers websites. These usually provide free access to on-line methods using their instrumentation, and also background information on the analytical techniques. Some of these require you to register your details, but normally the service is free.

Some of the better ones are:

- Varian
- Hewlett Packard (as Agilent)
- Supelco
- Perkin Elmer

Putting two halves together

Sometimes you may not be able to find exactly the analysis you are after in the literature, in that the exact combination of analyte and sample is not available. You will find references to the analyte in different samples, and other analytes in the sample you are interested in. Further, think about what other sample types may be similar in nature to yours: for example, if you are analysing biscuits, then flour or bread is a similar matrix.

However, it is important to remember when looking for information that in many analytical procedures, sample preparation is quite separate to the analysis step, as illustrated in Figure 1.2.

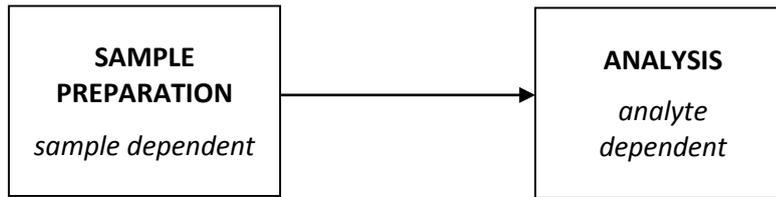


FIGURE 1.2 *The analytical process*

Sample preparation usually involves getting the sample into solution and at the right concentration levels. Though what is done will be partly determined by the analytical procedure to come (eg masking agents, pH correction), it is mostly related to how difficult the sample matrix is to break up. For example, analysing iron in iron ore is going to require much more effort in sample preparation than iron in creek water.

The *analytical* procedure is specific to the analyte; depending on matrix, the sample nature may be of no importance. Thus, once the iron ore is in solution, it may be analysed exactly the same as the creek water.

How does this help you? Look for both halves of the analytical task: the sample and the analyte. It may be that you can join two different analytical procedures together. You might find a perfectly good sample preparation step in a procedure which analyses a different analyte. This could be joined to a procedure which describes how to analyse a solution of your analyte, even though it has come from a different sample type.

EXERCISE 1.1

Your analytical task is iron in Corn Flakes, but you can't find an exact reference to this. What else could you look up?

Documenting your search

Whether you are writing a report (eg your project report) or doing a search to find a method to solve your problem, you should always record where you found the information, if for no other reason than being able to find it again.

In the case of a report, whenever you quote someone else's work in yours, you must acknowledge it in a reference list at the end of the report, and indicate where the work is referred to in the main body of the report. This is done in a particular, fairly standardised way, as shown below.

For a **journal article**, the order of listing is: author(s), journal title (underlined or in italics), volume, year and page numbers of article.

e.g. Smith FA, *Journal Of Mongolian Chemistry*, 69 (1969) 181-89.

For a reference from a **book**, the name of the journal is replaced by the title of the book, and there will be no volume number, but instead the book publisher.

e.g. Lo, WH & Phat, B, "Analysis Of Yak Products", Ulan Bator Press, 1981, p 69.

To indicate the use of this work in your introduction, procedure or discussion, there are two possible methods:

- the list of references is numbered, and the appropriate number is superscripted in the text where its reference is used, i.e. "... the level of unsaturated fatty acids in yak fat has been found to be 50 mg/100 g¹, though other studies ...", or
- the author's name and the date of the work is included in brackets in the text where the work is being referred to, i.e. "... yak meat is known to be high in iron (Lo & Phat, 1981), and also ...".

Either of these methods will be accepted in your project report. **It is not sufficient to include a list of information sources in a bibliography at the end of your report.**

1.2 Defining the task

Not all procedures will be suitable in practice even though they are, in principle, capable of analysing the desired analyte. There can be reasons for chemistry – e.g. matrix interferences – or logistical, e.g. the laboratory doesn't have the required instrument.

The simplest way to determine whether a procedure will be suitable, or which of a number of alternative is best, is to see how key characteristics of the task match with those of the procedure. The task is more than just the analyte and sample (eg sodium in beer), it is the nature of the sample (the what), and the conditions under which the analysis will take place (the who, when, where and how much).

The key factors involved in matching task and procedure are:

- sample concentration
- physical state of the sample
- complexity of sample matrix
- accuracy required
- number of analyses to be performed (per day and overall)
- time allowable for result
- multiple analytes
- existing laboratory equipment
- laboratory budget for purchases
- skill of technician assigned to task

EXERCISE 1.2

Complete the table below by explaining why each characteristic of the task is significant in the choice of the procedure. The first is completed as an example.

Sample concentration

- all techniques have a defined upper and lower limit of concentrations able to be analysed
- it is inaccurate and/or time-consuming to **substantially** dilute or concentrate a sample to make it fit to the procedure

Physical state of the sample

EXERCISE 1.2 (CONT'D)

Complexity of sample matrix

Accuracy required

Number of analyses to be performed

Time allowable for result

Multiple analytes

Existing laboratory equipment

Laboratory budget for purchases

Skill of technician assigned to task

Obviously you will find the answers to the characteristics of the task in the information provided to you and in your own laboratory situation. However, with the procedure(s), you need to have some knowledge of their own special characteristics.

Overview of analytical techniques

During this course, you have studied a large number of analytical techniques, and it is assumed that you are aware of the basic theory and practice of each. The purpose of these notes is to emphasise those aspects of the techniques with which you may not be as familiar:

- initial cost
- running costs
- accuracy
- time required
- operator skill levels

and remind you of the advantages and disadvantages of each technique. Some will be left for you to complete.

We will look at the techniques grouped by their concentration ranges (high, medium and low). Figure 1.3 gives a graphical overview of the typical concentration ranges for each technique listed below:

- | | |
|---|---------------------------------|
| • AAS – flame | • AAS – vapour generation |
| • AAS – electrothermal * | • Emission spectroscopy – flame |
| • Ion-selective electrodes | • Emission spectroscopy – ICP * |
| • Volumetric – indicator & instrumental | • ICP-mass spectrometry * |
| • UV-VIS absorption spectroscopy | • Infrared spectroscopy |
| • Molecular fluorescence * | • X-ray fluorescence * |
| • Polarography * | • ASV * |
| • HPLC | • GLC |

* indicates techniques covered in *Advanced Instrumental Techniques* (for Chem/Forensic Diploma students).

Methods For High Concentrations (1-100 g/L)

In general, the higher the concentration range (the lower the sensitivity), the more accurate and precise a technique is. This is not the case with XRF and spark emission, due to matrix effects.

Volumetric Analysis (indicator and instrumental)

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	Quick, simple and accurate, these are the best methods for high concentrations, but are not selective.

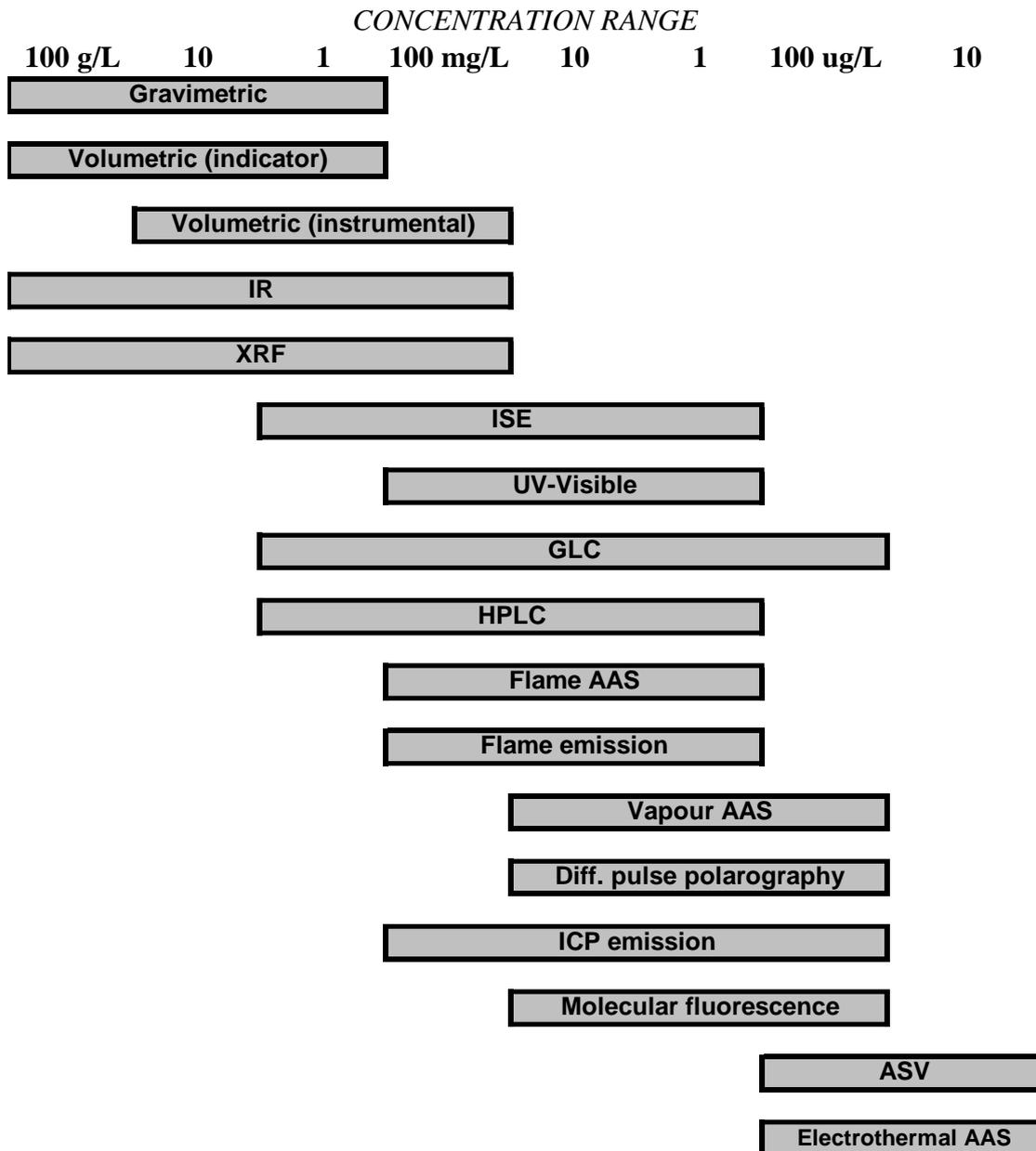


FIGURE 1.3 Classification of techniques by concentration range (see note below)

NOTE

ICP-Mass Spectrometry is not included in Figure 1.3, because it is a very new technique. Its working range is 100 ug/L to 10 ng/L.

Infrared Spectroscopy

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	Of limited application, due to the complexity and overlapping nature of component spectra in a mixture, FT-IR has improved the accuracy of the technique.

Methods For Medium Concentrations (1-100 mg/L)**X-Ray Fluorescence**

<i>Accuracy</i>	Limited by matrix effects.
<i>Time Required</i>	Low.
<i>Costs</i>	Very high initial (> \$100,000) moderate running costs
<i>Operator Skill</i>	High.
<i>Multi-component</i>	Yes.
<i>Overall Comments</i>	The ability to analyse solid samples for multiple analytes directly is a significant advantage, but the lack of accuracy and cost are significant disadvantages.

ICP Emission Spectroscopy

<i>Accuracy</i>	High, due to lack of interferences.
<i>Time Required</i>	Rapid.
<i>Costs</i>	> \$100,000 for purchase, and high running costs due to use of argon.
<i>Operator Skill</i>	Limited skills required, once computer software learnt.
<i>Multi-component</i>	Yes.
<i>Overall Comments</i>	The way of the future, particularly as prices decrease.

High Performance Liquid Chromatography

<i>Accuracy</i>	Moderate for instrumental technique, 3-5%.
<i>Time Required</i>	Relatively slow.
<i>Costs</i>	\$30-50,000 initial, moderate running costs.
<i>Operator Skill</i>	Very high.
<i>Multi-component</i>	Yes.
<i>Overall Comments</i>	Of unequalled versatility in the range of possible analytes, but not easy to use.

Differential Pulse Polarography

<i>Accuracy</i>	Matrix errors reduce accuracy, but typically 5%.
<i>Time Required</i>	Relatively slow.
<i>Costs</i>	\$5-40,000 initial depending on level of automation, moderate running costs, mainly involving mercury recycling.
<i>Operator Skill</i>	Medium.
<i>Multi-component</i>	Yes.
<i>Overall Comments</i>	The ability to analyse a range of analytes (ions and organic) at relatively low cost, and with relatively simple operations is somewhat offset by matrix errors and the generation of contaminated Hg.

Molecular Fluorescence Spectroscopy

<i>Accuracy</i>	5%, which at the 100 ug/L level is very good.
<i>Time Required</i>	Rapid.
<i>Costs</i>	\$5,000 for filter fluorimeter.
<i>Operator Skill</i>	Simple to use, more skill to set up a method.
<i>Multi-component</i>	Possible, but overlap of spectra causes errors.
<i>Overall Comments</i>	For strongly fluorescent species, the technique is superior to UV-VIS because of its sensitivity and wider linear range. A limited number of species are fluorescent.

UV-Visible Spectrophotometry

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	Good general purpose technique for a range of analytes, but may require obscure, expensive colour forming reagents for non-absorbing ionic species.

Flame Atomic Absorption Spectroscopy

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	The most commonly used technique for the analysis of low levels of metal ions, it is simple and accurate for most species, calcium and aluminium excepted!

Flame Emission Spectroscopy

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	The major application is the flame photometer, which is the best technique for Na and K - very simple, quick and reasonably accurate. An AAS can be used for the other metals, but is not really particularly satisfactory.

Ion Selective Electrodes

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	Quick and portable, but in most cases, of limited accuracy.

Gas Chromatography

<i>Accuracy</i>	
<i>Time Required</i>	
<i>Costs</i>	
<i>Operator Skill</i>	
<i>Multi-component</i>	
<i>Overall Comments</i>	With the advent of better columns and MS detection, a most powerful technique for the analysis of complex mixtures. Fewer operating difficulties than HPLC.

Methods For Low Concentrations (ug/L levels)**Vapour Generation AAS**

<i>Accuracy</i>	5-10%, which is acceptable for such concentrations.
<i>Time Required</i>	Slow.
<i>Costs</i>	\$5,000 initial outlay for attachment to normal AAS, moderate running costs.
<i>Operator Skill</i>	High.
<i>Multi-component</i>	No.
<i>Overall Comments</i>	Of most use for mercury, but complexity of setup means it is being superseded by ICP-MS.

Electrothermal AAS

<i>Accuracy</i>	5-10%, which is acceptable for such concentrations.
<i>Time Required</i>	Slow, but autosampler systems reduce operator time.
<i>Costs</i>	\$75-100,000 initial, moderate running costs.
<i>Operator Skill</i>	High.
<i>Multi-component</i>	No.
<i>Overall Comments</i>	Of wider application than ASV (the main alternative for ultra-trace metals) EAAS is harder to use and more expensive, but less prone to matrix errors. Being replaced by the more expensive but less problematic ICP-MS.

Anodic Stripping Voltammetry

<i>Accuracy</i>	5-10%.
<i>Time Required</i>	Slow.
<i>Costs</i>	\$40,000 for automated instrument.
<i>Operator Skill</i>	High, due to necessity for careful solution preparation.
<i>Multi-component</i>	Yes.
<i>Overall Comments</i>	For the small range of metals possible, it is superior to EAAS, since multi-component analysis is possible. Matrix errors are significant.

ICP-Mass Spectrometry

<i>Accuracy</i>	2-5%.
<i>Time Required</i>	Fast.
<i>Costs</i>	\$300-400,000
<i>Operator Skill</i>	High, due to necessity for careful solution preparation & operational requirements
<i>Multi-component</i>	Yes.
<i>Overall Comments</i>	The new "go-to" technique for ultra-trace analysis; very expensive but superior to the alternatives

EXAMPLE 1.1

The analysis is sodium in beer, with an expected concentration of 50-100 mg/L and a matrix containing potassium, calcium and magnesium at similar levels to analyte, 5% ethanol and a range of organics, including sugars.

The task characteristics are:

- maximum of 5% accuracy acceptable
- up to 50 samples/day to be analysed
- the work will continue for at least 12 months
- results are needed for QC so within 30 minutes of receiving the sample
- the laboratory has an AAS, but no sodium lamp, and no flame photometer
- a technician with 12 months experience will be doing the work

The proposed procedures use flame photometry (FP) and flame AAS (FAAS). The question is: which is the better option?

One way to answer this is to identify, for each factor, which alternative is superior. If one has more positives than the other, it is probably the better choice, though you should not treat this way as absolute, as one positive for method A might outweigh three positives for method B.

concentration	both require dilution, less for FP	Even
physical state	both require solution – equivalent	Even
matrix	FP less matrix affected, but needs internal std	FP
accuracy	similar	Even
number of analyses	both would cope with the expected workload	Even
time	FP quicker to set up, equal in running solutions	FP
multiple analytes	not relevant	n/a
existing equipment	FAAS already exists	AA
budget	FP \$5000, new HCL \$500	AA
skill	FP simpler	FP

So there are positives for both sides, FP having more, but requiring an extra expenditure of \$4500. If the FAAS was being already being used a lot for other work, purchasing a FP would be the better choice; if the AAS was under-used, it would be the choice.

EXERCISE 1.3

Evaluate the alternative techniques for the analysis of:

(a) ethanoic acid in white and brown vinegar by GC or titration.

The expected concentration is 40 g/L and the matrix is fairly minimal. Required accuracy is 5%, since the acid content is a legal reporting requirement for vinegars. At most 5 samples per day are required, and are needed within 12 hours. The laboratory technician has 5 years experience, and the laboratory has only basic equipment.

No words are needed in the comparison table: simply put a tick or cross in the relevant column to indicate that the proposed instrument is suitable or unsuitable for the task. If it is particularly good or bad, you could put two ticks or crosses.

	GC	Titration
concentration		
physical state		
matrix		
accuracy		
number of analyses		
time		
multiple analytes		
existing equipment		
budget		
skill		

Which is the better option?

(b) nitrate in soil by HPLC or ion-selective electrode

The expected concentration is 10-500 mg/kg and the matrix is obviously complex with many other ions present. The purpose of the testing is to identify areas where there is an excess or shortfall of available N and therefore speed is more important than an exact value. Sampling and testing is done by the same person, and up to 30 samples are taken from any one farm. The technician has 3 years experience, and the laboratory has all the necessary equipment.

	HPLC	ISE
concentration		
physical state		
matrix		
accuracy		
number of analyses		
time		
multiple analytes		
existing equipment		
budget		
skill		

Which is the better option?

EXERCISE 1.3 (c)

Your laboratory gains a 12 month contract to monitor low levels (10-500 ug/L) of Pb, Cu & Zn in a creek near a mine. This will require the purchase of a new instrument, as your laboratory doesn't have anything capable. The sample load is likely to be relatively low (5/day) and results are required to be available within 24 hours. The three instruments proposed are ASV, EAAS & ICP-MS. The technician given the job will be the most experienced in the laboratory.

	ASV	EAAS	ICP-MS
concentration			
physical state			
matrix			
accuracy			
number of analyses			
time			
multiple analytes			
existing equipment			
budget			
skill			

Rank the options in order, and explain your reasons.

1 (best)	
2	
3	

What You Need To Be Able To Do

- describe the process for developing an analytical procedure
- define an analytical task
- compare analytical techniques
- evaluate alternative procedures