Headaches, Colds and Cures

A group experiment in chemical analysis

North East regional heat
Schools Analyst Competition 2012

Royal Society of Chemistry, Analytical Division

Headaches, Colds and Cures

Today's competition is to analyse a number of commonly used pharmaceutical preparations……. taken everyday by thousands.

There are three exercises for each team to do :

1. Analysis of aspirin in tablets by titration
2. Analysis of tri-sodium citrate in Boots Cold & Flu relief
3. Analysis of paracetamol in tablets by UV spectrometry

None of the exercises should take one person more than 2 hours so this should leave plenty of time for calculations and answering the questions in the three hours of the competition. The competition is taking place in several other centres in the North East and Yorkshire and so we must keep to the time schedule to be fair to all the centres.

At the start of the exercise each team will be given 4 identical answer-books
- one for each team member to be used as a laboratory notebook
- one to be the team report to be handed in for marking

The first task will be to draw out a plan showing how you intend to divide up the tasks amongst the team members.

You will quickly see that two of the analyses will involve the use of instruments. Follow the instructions given to you on the availability of the instruments. Few, if any, university laboratories would have sufficient instruments if every team decided to use the same instruments at once. Plan your work so that the use of equipment is spread through the available time. Similarly you should structure your work so that the weighings are not all timetabled at the start of the exercise to avoid queuing at the balances. A careful look at the schedules will show that there are several tasks you can do before you even need to use the balances.

You should have read the section 'Background to the Instruments' found later in this booklet. A good analyst must know the theoretical background to an instrument and should not just blindly write down the results produced by the instrument or computer.

Note that in your work-book there are a number of questions which may be used in the marking as tie-breakers. Remember to allow time in your work plan for answering these questions as well as completing the workbook to be handed in.

What's in a tablet (or powder) ?

When you go to the chemist's shop and get your aspirin or phensic many people would think they are getting a pure drug. It is never the case. Sometimes the drug is the major portion of the tablet but more often it is only a minor part. There are many other ingredients (excipients). These include

Adhesives/binders eg gelatin, starch, polyethylene glycols

Diluents eg calcium phosphate, lactose, sodium sulphate

Disintegrating and wetting agents eg agar, bentonite, sodium lauryl sulphate

Flavouring agents eg cocoa, liquorice powder, sodium citrate.

None have any direct physiological effect but they are essential in getting the correct dosage of drug delivered to its point of action and in a form which people don't mind taking.
The specification of any tablet will include the nominal mass of active ingredient in a tablet and also the permitted deviation from the nominal mass. Don't expect the analyses you are undertaking to show 100% drug.

Today's experiments are to measure the ingredients in three common over-the counter remedies for headaches and colds, namely aspirin, paracetamol and Lemsip. Each of these has ingredients belonging to a class of drugs known as analgesics. Methods are modified from the authoritative guide in Britain to drug analysis – the British Pharmacopoea (BP). One of the tie –break questions asks you to calculate whether the drug is within the permitted range from the stated amount as given in the BP.

Analgesics

Drugs commonly used to relieve pain

Safety !

Safety is of utmost importance in the laboratory

- At all times wear buttoned laboratory coats and safety spectacles in the laboratory.
- There should be no eating, drinking or chewing of gum in the laboratory.
- Always use the pipette fillers provided. You should never mouth-pipette.
- As with all laboratory procedures at the university a safety assessment will have been made of the exercises and will be available in the laboratory. Major points from these will be emphasised in the talk at the start of the exercise.
- If there is a fire alarm, leave what you are performing immediately and walk calmly out of the building. Do not spend time picking up your coat and belongings. Do not use the lift.
- There will be first aid-trained personnel present in the laboratory. These will be pointed out to you by the demonstrators.

Note also that the exercise is not finished until the apparatus is left clean and tidy (according to instructions that will be given to you in the laboratory) AND all compounds and solutions have been disposed of in a safe manner. Aqueous solutions can be flushed down the sink with running water.

IF YOU ARE IN DOUBT ABOUT ANYTHING ASK A DEMONSTRATOR

1. Analysis of Aspirin in Tablets by titration

Aspirin tablets comprise of acetylsalicylic acid in a neutral inert medium. The British Pharmacopoea (BP) analysis of these tablets to confirm their specification involves reacting the tablet with excess base. The excess base is then back titrated with standard acid.

Equipment

2x weighing boat/bottles
20 cm^3 pipette
10 cm^3 pipette
pasteur pipette
50 cm^3 burette
white tile
wash bottle containing deionised/distilled water
4 aspirin tablets
2 x 250 cm^3 conical flasks
2 x small funnel to fit flasks
pipette filler
2x bunsen burners, tripods, gauzes
0.5 M NaOH
0.5 M HCl (standardised)
mortar and pestle

The following procedure should be performed twice

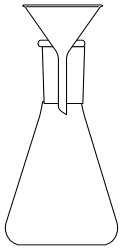
1.1 Crush 2 aspirin tablets using the mortar and pestle. Weigh approx 0.6 g aspirin tablet powder in a weighing boat/bottle on a laboratory balance. Transfer the weighing bottle to an analytical balance to obtain the accurate mass.

1.2 Tip the powder into a 250 cm^3 conical flask and reweigh the weighing bottle on the analytical balance. Calculate the accurate weight of powdered aspirin transferred.

1.3 Pipette 30 cm^3 0.5 M sodium hydroxide into the flask.

1.4 Pipette 30 cm^3 0.5 M sodium hydroxide into a second (empty) flask to act as a blank.

1.5 Place a small funnel on the top of each flask to prevent excessive evaporation of the water when heated.



1.6 Heat both flasks to boiling point then simmer gently for 10 min. If necessary add small amounts of de-ionised or distilled water to maintain the liquid level. The flask containing the aspirin will have become slightly yellow coloured, the blank remaining colourless.

1.7 Cool the flasks under running water. Add a few drops of phenol red indicator solution solution to each and titrate the excess alkali with standardised 0.5 M hydrochloric acid. The end point is red to the original yellow for the aspirin solution (titre x ml) and red to a

paler yellow for the blank (titre y ml). Note the exact concentration of the hydrochloric acid solution.

1.8 Calculate the volume of 0.5 M hydrochloric acid used in the reaction with the aspirin (y-x) cm^3. From this you will be able to calculate the number of moles of sodium hydroxide used in the reaction with the aspirin.

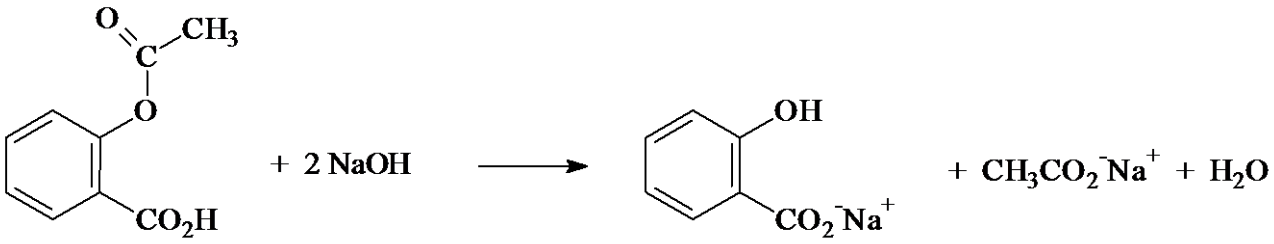
1.9 Calculate the mass of aspirin in the sample according to the method given in the workbook and using the equivalence

1 mole aspirin ≡ 2 moles sodium hydroxide

1.10 Calculate the percentage aspirin in the powder.

Reactions

$$NaOH + HCl \longrightarrow NaCl + H_2O$$



Aspirin

2. Analysis of tri-Sodium Citrate in Boots Cold & Flu Relief Powder

The tri-sodium citrate is included in the powder as part of the fruity taste. The active ingredients are ascorbic acid and the analgesic paracetamol.
After dissolution of the powder, the sodium concentration of the solution is determined by flame photometry. This is then converted into the concentration of tri-sodium citrate. From this the mass of tri-sodium citrate per 5 g Cold & Flu (approx weight of sachet) is calculated.

Equipment

Weighing boat/bottle
20 cm^3 pipette
10 cm^3 pipette
5 cm^3 pipette
pasteur pipette
wash bottle containing deionised/distilled water
250 cm^3 volumetric flasks
4 x 100 cm^3 volumetric flasks
small funnel
pipette filler
sample of Cold & Flu relief

2.1 Weigh approx 0.125g Cold & Flu powder in a weighing boat/bottle on a laboratory balance. Transfer the weighing boat/bottle to an analytical balance to obtain the accurate mass.

2.2 Tip the Cold & Flu powder into a 250 cm^3 volumetric flask using a funnel and reweigh the weighing bottle on the analytical balance. Calculate the accurate weight of Cold & Flu transferred.

2.3 Add de-ionised or distilled water to the flask, at the same time washing any remaining grains of Cold & Flu on the funnel into the flask. Dissolve the Cold & Flu by inversion and shaking the flask (volumetric flasks must NOT be heated) and make up to volume with de-ionised or distilled water.

2.4 A stock solution containing 100 mg dm^{-3} sodium (Na^+) is provided. From this make up a series of four standards over the range 0-20 mg dm^{-3} using the pipettes and volumetric flasks provided. You may need to perform the dilution in more than one step.

2.5 Take the standards, a blank (de-ionised or distilled water) and the sample solution to the flame photometer. A demonstrator will show you how to use the instrument. Aspirate the standards into the instrument, starting with the lowest standard first and noting the

readings. Then aspirate the sample solution. The reading of this should fall in the range of the standards.

2.6 Draw a calibration graph (emission intensity vs concentration) using the standard solutions and from this determine the concentration of sodium in the sample solution.

2.7 Given the formula of sodium citrate is $C_6H_5O_7Na_3$ and the method described in the workbook, calculate the concentration of sodium citrate in the solution (mg dm^{-3}).

2.8 From this calculate the mass of sodium citrate in the original sample of Cold & Flu. Finally convert this to the mass per 5g Cold & Flu powder.

3. Analysis of Paracetamol in tablets by UV Spectrometry

The analysis involves dissolution of the tablet, filtration to remove insoluble components, adjustment of the pH, then measuring the ultraviolet absorbance which detects and quantifies the aromatic ring in the paracetamol molecule. There have to be a number of dilutions due to the sensitivity of the technique.

Equipment

2 x weighing boats/bottles
100 cm^3 conical flask
2x 10 cm^3 pipettes
50 cm^3 measuring cylinder
pasteur pipette
wash bottle containing deionised/distilled water
1 paractemol tablet
2 x 250 cm^3 volumetric flasks
9x 100 cm^3 volumetric flasks
funnel + filter paper
pipette filler
0.1M NaOH
quartz 1cm cell for uv
mortar and pestle

3.1 Crush the tablet using the mortar and pestle and grind until a fine powder is obtained. Weigh approx 0.21 g paracetamol tablet powder in a weighing boat/bottle on a laboratory balance. Transfer the weighing boat/bottle to an analytical balance to obtain the accurate mass.

3.2 Tip the tablet powder into a 250 cm^3 volumetric flask using a funnel and reweigh the weighing boat/bottle on the same analytical balance. Calculate the accurate weight of paracetamol powder transferred.

3.3 Add approx 50 cm^3 0.1M sodium hydroxide using a measuring cylinder to the flask, at the same time washing any remaining grains of tablet powder on the funnel into the flask. Add approximately 50 cm^3 water and place in an ultrasonic bath for 1 min. If no ultrasonic bath is available the flask contents may be mixed thoroughly by repeated inversion and shaking for 2 minutes.

3.4 Make up to 250 cm^3 with deionised water and mix.

3.5 Filter 25-50 cm^3 of the solution using a filter funnel and filter paper. Pipette 10 cm^3 of this filtrate into a 100 cm^3 graduated flask, make up to volume with deionised or distilled water and mix.

3.6 Pipette 10 cm^3 of the solution into a 100 cm^3 volumetric flask, pipette in 10 cm^3 0.1M sodium hydroxide, make up to volume with deionised or distilled water and mix. . This is your unknown solution.

3.7 Repeat steps 3.1 – 3.5 with 0.19 g powdered paracetamol standard. **Try to be as close as possible to this value and within the range 0.189-0.191.**

3.8 Pipette 0*, 5, 10, 15, 20 cm^3 of the solution from step 3.7 into five 100 cm^3 volumetric flasks, pipette in 10 cm^3 0.1M sodium hydroxide, make up to volume with deionised or distilled water and mix. These are your calibration solutions and represent masses of 0, 0.075, 0.15, 0.225, 0.3 g paracetamol in the original mixture.
* this simply means don't do anything at this stage but you do add the sodium hydroxide and water later.

3.9 Measure the absorbance of the unknown and calibration solutions at 257 nm using quartz cells, blanking with the zero calibration solution (the demonstrator will explain how to do this). "Blanking" means setting the instrument to read zero when there is no unknown present.

3.10 Plot a calibration graph of absorbance vs mass of paracetamol. From this read the mass of paracetamol in the sample and so determine the percentage paracetamol in the powder.

Are all analgesics the same?

If you have a headache you will probably just reach for the first bottle of tablets you come across whether they are aspirin, paracetamol or any other analgesic. They are not all the same:-

If you are a blood donor you will know that the effects of aspirin last for several weeks and so you are not allowed to give blood for a month after you have taken aspirin. Paracetamol breaks down more quickly and you can give blood within a week.

If you have tinnitus (a continuous noise in the ear which affects about 10% of the population in the UK) you will know that aspirin aggravates the problem, paracetamol having no effect.

Paracetamol in quantiites greater than the stated dose can have disasterous effect which can cause a painful death. Aspirin is a more forgiving compound, causing less sever effects if taken too liberally, but still causing death in a high dose.

Aspirin in lowish doses has secondary beneficial effects such as protection against heart disease.

Background to the Methods and Instruments used

The Beer-Lambert Law

Many compounds which we see in the world are coloured. We see the colour because when white light shines on to the material some of it is absorbed and the rest is reflected to our eyes. The colour which we see is that which is NOT absorbed. Remind yourselves what colours are found in the visible region of the spectrum

ROYGBIV

If a material looks red it is because it is absorbing all the other components of the spectrum

OYGBIV

Similarly if something is blue it is absorbing

ROYG_IV

The absorption of light is measured by a spectrometer which shines light at a specific wavelength through a solution and measures the amount of light passing through the solution. The reading it produces is the absorbance of the solution and is defined as

$$\text{Absorbance} = \log (I_o / I)$$

Where I_o = intensity of incident light
I = intensity of light after passing through the solution

If the solution is sufficiently dilute, the absorbance of the solution at a specific wavelength is proportional to its concentration. This is known as the Beer-Lambert law or sometimes simply Beer's law.

$$A= \varepsilon.c.l$$

Where A = absorbance
ε = constant
l = pathlength of absorbing solution

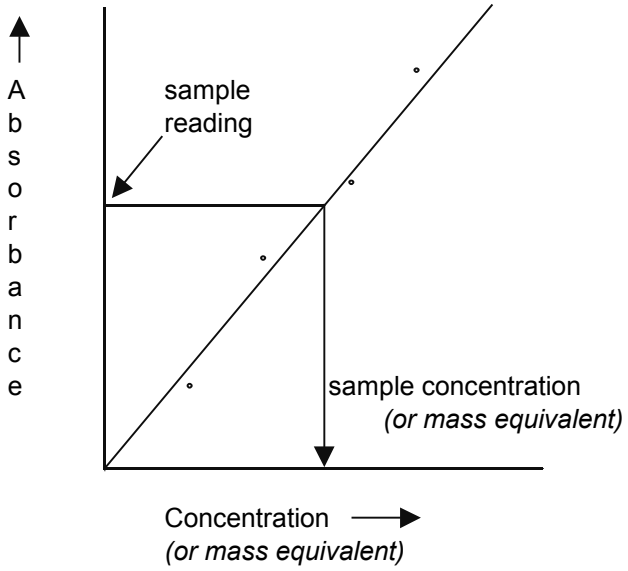
The law applies through the whole electromagnetic spectrum. Today we will be measuring the absorption of ultraviolet light.

In order to determine the concentration of a compound which absorbs light you simply make up a number of standard solutions of gradually increasing concentration and plot a graph of absorbance vs concentration. This should be a straight line passing through the origin. If you then measure the absorbance of the unknown solution you can determine the concentration from the calibration graph (Figure1).

In order to simplify the calculations today we are replacing the concentrations on the x-axis with masses of standards. You might want to check when you return home that this will produce exactly the same result but without several lines of maths!

Figure 1

Beer-Lambert law calibration graph



Flame Photometry

Quite often if you put a material into a flame, the flame becomes coloured. In many instance the colour is due to a particular element in the flame. If you have never have seen this at home or demonstrated in a laboratory you will certainly have seen fireworks where each of the (literally) flamboyant colours is produced by different elements being heated to the point that they give off light. If the element is introduced to the flame in solution, the intensity of light which is given off is proportional to the concentration of the element in solution. Take care not to say this is the Beer-Lambert law. The Beer-Lambert law is concerned with the absorption NOT emission of radiation.

This technique is used in a whole range of instruments ranging form one of the simplest analytical instruments in the laboratory (flame photometer – Figure 2) to one of the most sophisticated (inductively coupled plasma-optical emission spectrometers).

The calibration plot of emission intensity vs. concentration is normally a straight line. However occasionally the method uses requires calibration slightly above the linear range and in this case the line may have a slight curvature.

Figure 2

A Flame Photometer