Learn Chemistry

Problem Based Practical Activities

Problem 10: Patient prognosis

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Problem 10:
Patient prognosis

Curriculum links;
transition metal complexes, colorimetry, alcohols, carboxylic acids, esters, analytical techniques

Practical skills;
dilution, colorimetry, observation skills, GC analysis

A nineteen year old male has recently collapsed. His doctor would like the students to test;

i) the patient's urine for glucose
ii) the concentration of salicyclic acid (the break down product from aspirin) in the patient's urine [by colorimetry of the iron (III) salicylate complex]
iii) the patient's blood alcohol level (by interpretation of GC's provided)

Using this information the students are asked to make a recommendation as to the reason why the patient fainted.

Extension discussion points:

- Why are solutions of iron(III) ions more acidic than ethanoic acid? How does this help to explain the formation of the $[Fe(H_2O)_4(salicylate)]^+$ complex?
- What should be used for the blank in the colorimetry experiment?

Problem 10: Patient prognosis

Pre-Lab questions

(Remember to give full references for any information beyond A-level that you find out)

1. Glucose is a reducing sugar.

a) Draw the structure of glucose

b) Use your knowledge of chemical tests in biology or chemistry to indicate a test you could use to show the presence of glucose in an unknown sample

c) Why is the presence of glucose in a patient's urine a cause for concern?

2. Aspirin is one of the most widely used painkillers in the world today. It works by inhibiting the formation of prostaglandins which are the chemicals responsible for the sensitisation of nerve endings.

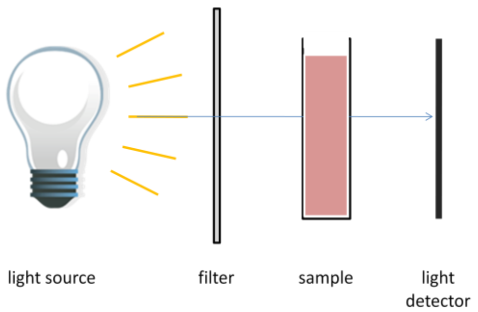
a) Draw the structure of aspirin and highlight the functional groups that you recognise.

b) In the digestive tract, the aspirin is hydrolysed under basic conditions to form salicyclic acid and ethanoic acid, before being absorbed into the blood stream. Write an equation for the hydrolysis reaction.

c) The reaction in fact occurs under mildly alkaline conditions in the digestive tract. Draw the structure of salicylic acid under these conditions.

3. Gas chromatography or GC is an analytical technique that can be used to identify unknown substances in a sample. Explain how GC is used to test athlete's blood or urine for drug taking. How is the process quantitative?

4. Colorimetry is an analytical technique that can be used to determine the concentration of a coloured compound in a solution by measuring the absorbance of light by the sample relative to a sample of the same substance of known concentration. A simple diagram of a colorimeter is shown opposite;



a) Explain the purpose of the filter. How is the appropriate filter for a solution chosen?

b) Salicyclic acid can be detected in a urine sample by complexation of the salicylate anion with Fe^{3+} ions. A six co-ordinate complex, $[Fe(salicylate)(H_2O)_4]^+$ is formed. This complex absorbs light at 520 nm and therefore appears to be pink/purple in colour.

i. Draw the structure of the complex formed.

ii. What colour of filter should be fitted when using a colorimeter to record the absorbance of a solution of this complex?

Problem 10: Patient Prognosis

Introduction

Martin's medical practice

Larchester

Dear chemist,

I need your help with diagnosing a patient who recently reported to me after fainting during an evening out with friends. The patient is a nineteen year old male, with a BMI of 27.5 and blood pressure of 110 / 72. The patient reports suffering from a severe headache prior to his night out for which he took aspirin.

The patient is a keen rugby player with hopes of performing at international level. It is therefore important that we find the underlying cause of the fainting as soon as possible. In the first instance, I would like to rule out excess alcohol intake, diabetes or accidental aspirin overdose as the cause. Signs and symptoms of aspirin toxicity begin to appear at salicylate levels of higher than 300 mg/L in the patient's urine.

Accompanying this letter, you will find a urine sample and a GC analysis of the patient's blood. These were obtained immediately after the patient regained consciousness. Please use these to provide;

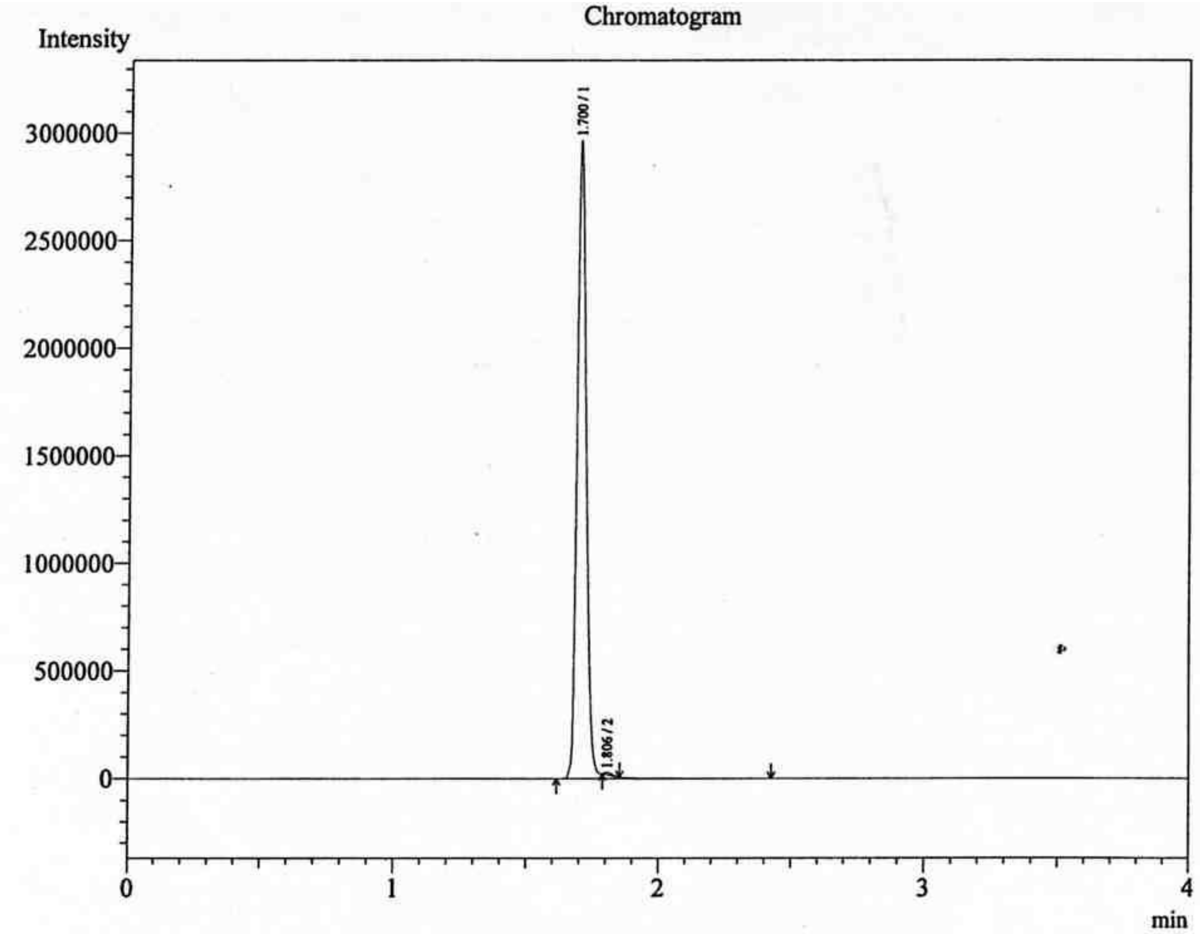
1. A qualitative test for the presence of glucose in the patient's urine
2. A quantitative assessment of the patient's blood alcohol content (BAC)
3. A quantitative assessment of the level of salicylate (the waste product from aspirin hydrolysis) in the patient's urine

The level of salicylate in the urine can be quantified by colorimetry. Complexing 1 cm^3 of the solution containing salicylate with 4 cm^3 of a 5% iron (III) chloride solution will give a coloured solution, the absorbance of which can be calibrated against a known concentration of salicylate. A solution of salicylate of concentration 500 mg dm^{-3} has been provided. This can be diluted to known concentrations for calibration.

Please provide a full report detailing the results of your group's analyses. For each analysis indicate whether the result is as would be expected. Make a recommendation, with reasoning, as to the cause of my patient's fainting.

Kind regards,

GC Analysis of Patient's Blood Sample

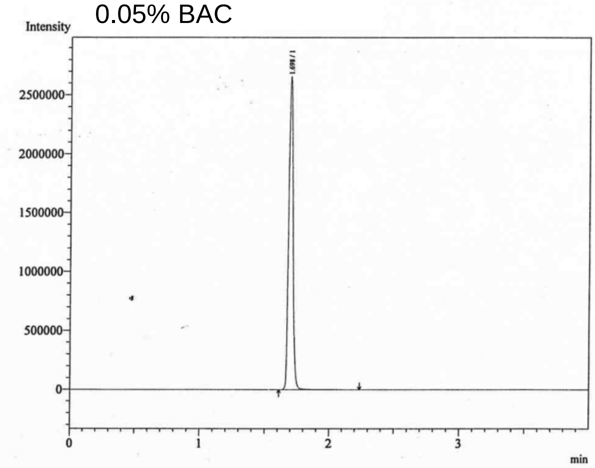


Peak Table - Channel 1

Peak#	Ret.Time	Height	Area	Area%
1	1.700	2931722	7169868	99.7271
2	1.806	10563	19623	0.2729
Total		2942285	7189491	

Chromatograms of blood samples of known alcohol content

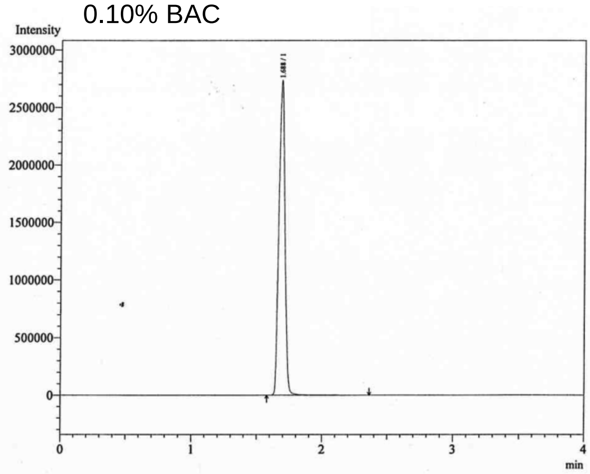
0.05% BAC



Peak Table - Channel 1

Peak#	Ret.Time	Height	Area	Area%
1	1.698	2642834	6242476	100.0000
Total		2642834	6242476	

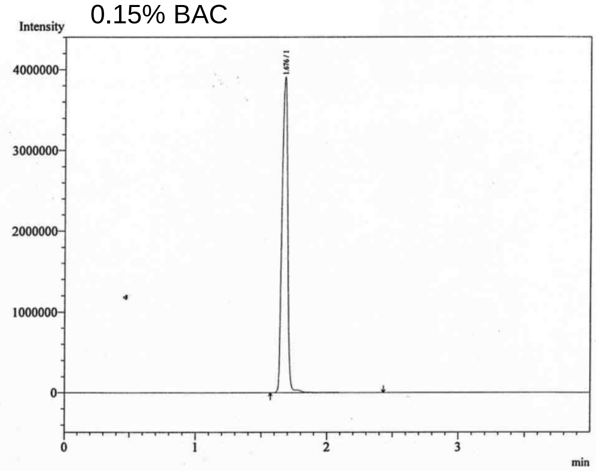
0.10% BAC



Peak Table - Channel 1

Peak#	Ret.Time	Height	Area	Area%
1	1.688	2737045	8768689	100.0000
Total		2737045	8768689	

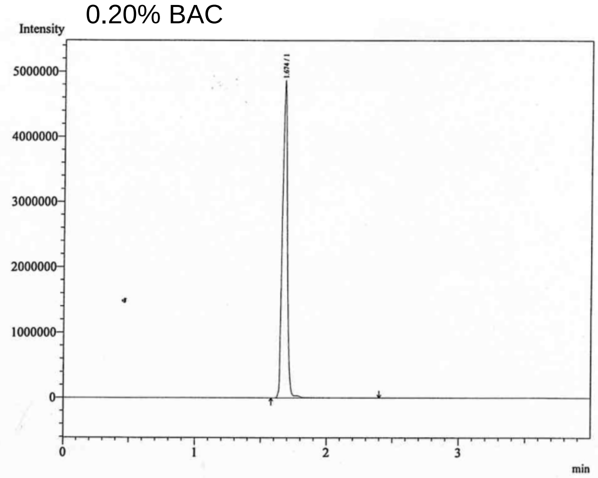
0.15% BAC



Peak Table - Channel 1

Peak#	Ret.Time	Height	Area	Area%
1	1.676	3890110	11738282	100.0000
Total		3890110	11738282	

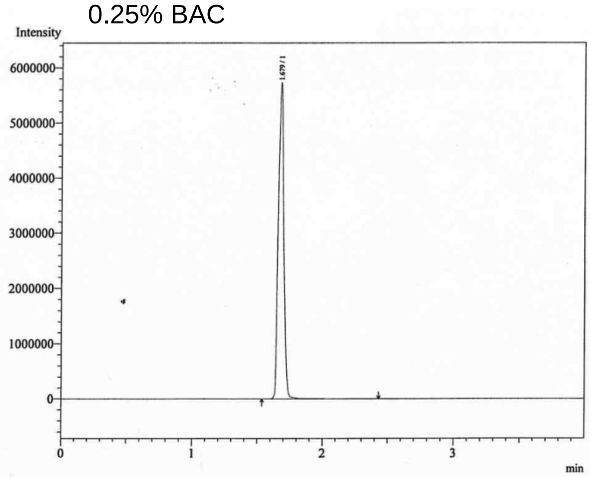
0.20% BAC



Peak Table - Channel 1

Peak#	Ret.Time	Height	Area	Area%
1	1.674	4852348	14155977	100.0000
Total		4852348	14155977	

0.25% BAC



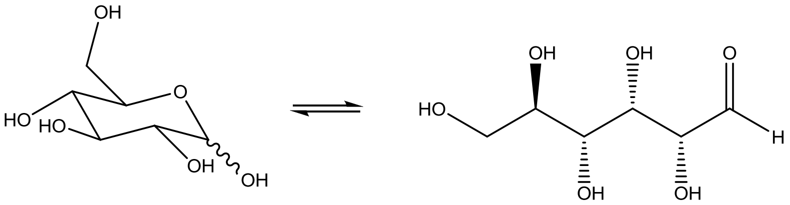
Peak Table - Channel 1

Peak#	Ret.Time	Height	Area	Area%
1	1.679	5718831	16714044	100.0000
Total		5718831	16714044	

Teacher and Technician Pack

Pre-Lab answers

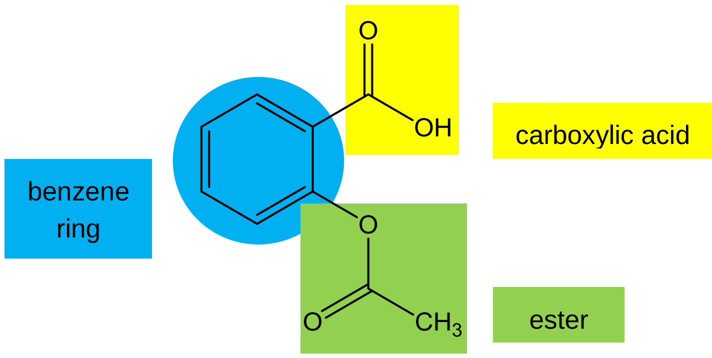
1. a)



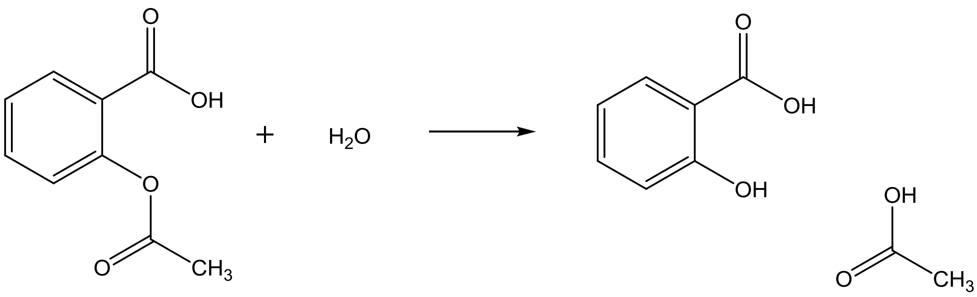
b) Any test for an aldehyde will indicate the presence of a reducing sugar. The most common test is Benedict's or Fehling's test where heating a small amount of the sample with either Benedict's or Fehling's solution brings about a change in colour from blue to brick red.

c) The presence of glucose in a patient's urine is a concern as it can be a sign of diabetes. Normally, urine contains no glucose because the kidneys are able to reclaim all of the filtered glucose back into the bloodstream. Presence of glucose in the urine indicates high blood glucose levels which may be caused by diabetes.

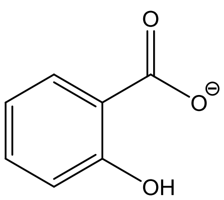
2. a)



b)



c)

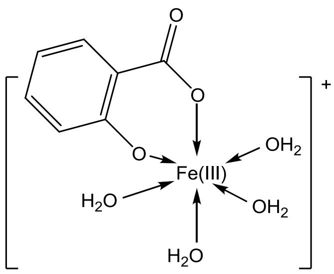


3. In **gas chromatography**, the sample is vaporized in the presence of a gaseous solvent (the mobile phase) and passed through a long capillary tube packed with a powder coated with oil (the stationary phase). Each substance moves along in the gas / oil mixture differently and stays in the gas phase for a unique, specific time, called the **retention time,** before exiting the end of the column and being detected. The retention time can be used to identify the compound present in the sample by comparison with the retention times of known drugs.

The results are presented on a graph or chromatogram consisting of one or more peaks for each of the different components in the sample. The technique is quantitative because the **area under the peak is proportional to the amount of that component present**. Thus by comparison with calibrated samples containing known quantities of the identified drug, the quantity of that drug in the sample can be determined.

4. a) The filter is chosen such that the light it transmits is absorbed by the sample. It selects the wavelength of visible light which is transmitted e.g. a blue-green filter allows blue-green light (wavelength 500-590 nm) only through. So if a sample appears red this means that it absorbs light of the complimentary colour to red i.e. blue-green. Hence a blue-green filter must be chosen.

b)



Complex colour: pink-purple

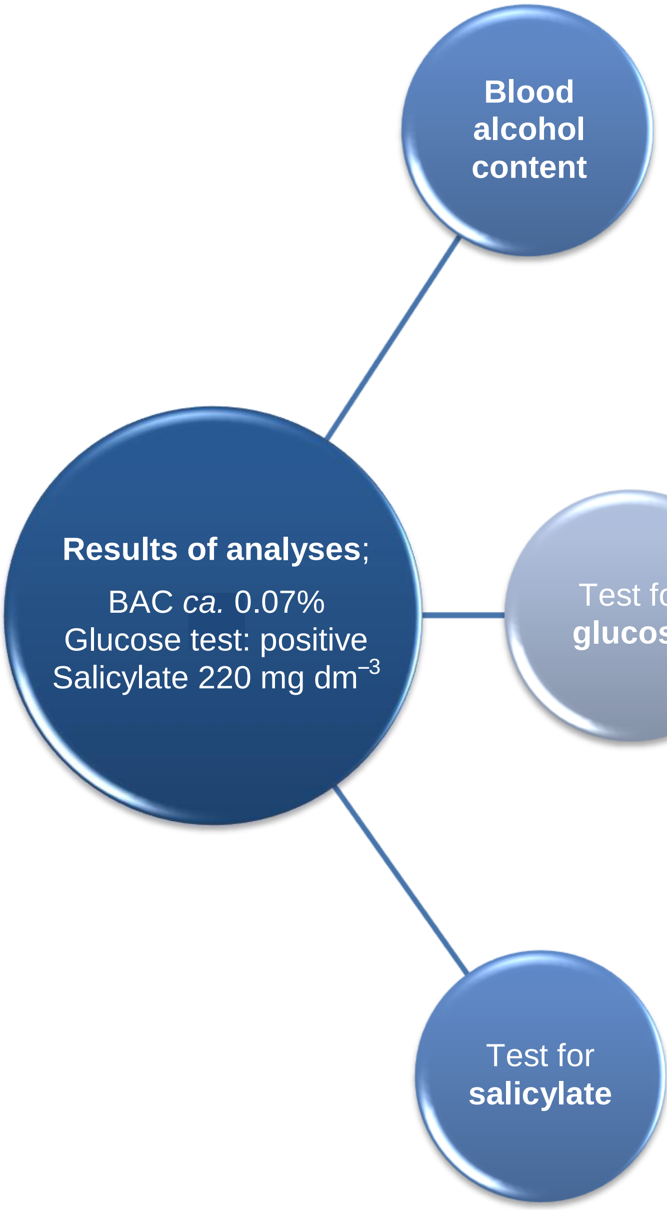
Filter colour chosen: blue-green

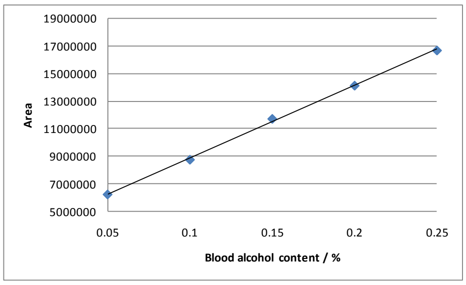
(If the students ask the equation for the formation of the complex is;

$[Fe(H_2O)_6]^{3+}$ + salicylate$^-$ $\rightarrow$ $[Fe(H_2O)_4(salicylate)]^+$ + 2 H_2O + H^+

The iron(III) ion is sufficiently polarising as a result of its large charge to size ratio for the phenolic OH to be deprotonated)

Proposed method





- Students produce a calibration graph from the GC traces provided
- The students then use the calibration graph and the GC trace of the patient's blood to determine the BAC of the patient

Students identify the presence of glucose in the patient's urine using the Benedict's test;

- Place a small amount of the urine sample in a test tube
- Add Benedict's solution and heat gently in a water bath
- A change of colour from blue to brick red indicates the presence of a reducing sugar, in this case glucose

Alternatively, students can test the urine with Clinistix®

Students determine the concentration of salicylate in the urine sample as indicated below;

- Make up a solution of 1 cm^3 of water and 4 cm^3 of 5% iron(III) chloride **[Irritant]** to use as a blank in the colorimeter
- Dilute the 500 mg dm^{-3} stock solution of salicylate **[Harmful]** to make solutions of concentration 100, 200, 300, 400 and 500 mg dm^{-3}.
- Place 1 cm^3 of each solution into a test tube and add 4 cm^3 of 5% iron(III) chloride solution.
- Transfer each new solution to a cuvette and measure the absorbance in the visible region from 400-600 nm.
- Repeat steps 2 and 3 above with the urine sample
- Use the absorbance to determine the concentration of salicylate in the patient's urine.

Recommendation: The patient's BMI falls into the overweight category. His blood pressure is normal. The blood alcohol level (approx. 0.067%) indicates that the patient was mildly inebriated on the night. The levels of salicylate (220 mg dm^{-3}) indicate that the patient had administered a therapeutic level of aspirin consistent with taking two aspirin for a headache. The presence of glucose in the urine is a concern and suggests the possibility of diabetes. Further investigations should be carried out in this area.

Teacher and Technician Pack

Equipment list

See the **Health and safety guidance** section in the **Introduction** for more general information on risk assessments and a key to the health and safety symbols used.

Each group will need;

A urine sample made up of;

12 mg sodium salicylate **[Harmful; Irritant]**

100 mg glucose **[Low hazard]**

5 drops of ethanol **[Highly flammable]**

Made up to 50 cm^3 with dilute cold tea (to create a "urine" look)

For the glucose test;

Test tube

Test tube holder

Disposable pipettes

Access to Benedict's solution

250 cm^3 beaker, Bunsen, tripod and gauze for a hot water bath

Or Access to Clinistix®

For the colorimetry;

Access to a colorimeter

6 × plastic cuvettes

50 cm^3 of a 5% by mass solution of iron(III) chloride **[Irritant]** (made by dissolving 8.32 g of $FeCl_3 \cdot 6H_2O$ in **WATER†** and making up to 50 cm^3)

An accurate method of measuring 1 cm^3 and 4 cm^3 (either 2 × 5 cm^3 plastic syringe or 2 × 10 cm^3 measuring cylinder)

Disposable pipettes

A 500 mg dm^{-3} stock solution of salicylate **[Harmful]** (made by dissolving 584 mg of sodium salicylate in 1 dm^3 of water). 50 cm^3 per group should be sufficient. This can be placed in a burette to facilitate the dispensing of small quantities.

Equipment for accurate dilution of the stock solution (e.g. a burette of distilled water and small test tubes and bungs)

Distilled water

Test tube rack

† **NOTE** If the iron (III) chloride solution is made up in acid as recommended by CLEAPSS it protonates the sodium salicylate and no complex forms. It must therefore be made up **IN WATER**.