WMI

The origins and development of the modern pharmaceutical industry

In this activity your class will divide into three groups. Each group will prepare a short presentation on one of the following topics.

1 Folklore in medicine
2 Medicine discovery and the nation's health
3 The British pharmaceutical industry

Finding information

You are expected to research your topic by searching the Internet. Start by looking at the Salters Advanced Chemistry web site which will link you to other sites which are relevant. Some suggestions for the focus of your research and how you might organise your presentation are given below for each topic.

1: Folklore in medicine

A suggested format for your presentation is a discussion between two people, one arguing the case based on your research, for traditional remedies, the other the case of someone who does not believe in folk medicine.

The case *against* might include some of the following points.

- We are civilised now: we don't believe in medicine men, voodoo and superstition.
- Most of these medicines work by 'the placebo effect' – people *believe* the medicines are going to make them better, so they do. Take away the primitive beliefs and the medicines won't work.
- In primitive communities it was kill or cure. Any medicine was better than none.

2: Medicine discovery and the nation's health

Your group, in addition to searching the Internet, will look at part of the Association of the British Pharmaceutical Industry's publication *an A to Z of British Medicines Research*, given below and in Figure 1, and at a table of deaths from various diseases since 1935 (Table 1).

Here are some of the questions you might like to consider in your research and presentation.

- Is there a link between trends in deaths from disease and the discovery of medicines?
- What other causes of longevity are there as well as improvement of medicines?
- What are the main killers today? What will happen when they are tamed?
- Is death rate a good measure of the nation's health? What other measures are there?

Extract from *an A to Z of British Medicines Research*

The past fifty years of British medicines research

The past 50 years have seen a revolution in our understanding of disease and its treatment (Figure 1). It has been a half century in which:

- organ transplantation has become accepted as commonplace
- the structure of DNA was elucidated, opening the way for the sciences of genetic engineering and genomics
- interferon, the first of a family of naturally occurring molecules called cytokines, was isolated and named
- monoclonal antibodies were discovered, opening the way for the specific targeting of medicines
- smallpox, a centuries-old scourge of mankind, was eradicated from the planet

- techniques for *in vitro* fertilisation were developed, bringing family joy to many childless couples
- AIDS emerged and with it the discovery of a family of human viral pathogens called retroviruses, of which HIV is one, leading to entirely new medicines to contain the virus
- advanced scanning techniques were developed which have transformed the diagnosis and understanding of many human diseases.

Built on these and many other developments, there has also been a revolution in the number, specificity and safety of human medicines. It is here as much as anywhere that Britain has been in the front line: for the whole of the last half century, medical and medicines research in Britain has been the envy of the world for its innovative skills and high ethical standards. Even today, despite increasing pressure on funding and demands for greater safety in medical products, we still enjoy this reputation. British hospitals are often a first choice for assessing the value of new medicines. Many European, Japanese and American owned pharmaceutical companies choose to fund major research groups here. It is no accident that today 7 of the top 25 medicines in the world, and 6 of the top 12 medicines used to treat bacterial and viral infections, are the products of British pharmaceutical research.

But the industry can also be proud of the contribution it makes to the British economy. UK earnings from the export of medicines exceeded imports by £2.3 billion in 1997. On top of that, the industry employs the skills of more than 60 000 people, including 20 000 highly-trained scientists and doctors.

Disease	1935	1945	1955	1965	1975	1985	1995
Scarlet fever	499	82	21	1	–	–	–
Whooping cough	1473	689	88	21	12	4	2
Diphtheria	3408	694	13	–	1	–	1
Measles	1264	728	176	115	16	11	1
Typhoid	170	44	15	8	1	–	–
Tuberculosis (respiratory)	23 840	19 668	5837	2008	722	408	355
Syphilis	3521	2378	1385	857	110	40	9
Heart disease (circulatory system)	103 613	127 969	138 313	151 718	299 669	287 054	241 871
Cancer	62 602	73 753	91 340	106 474	123 769	141 618	140 791
Total deaths	477 401	481 274	518 864	549 379	582 841	590 734	565 902
Population (million)	40.65	42.63	44.44	47.67	49.47	49.99	51.52

Table 1 Deaths from various diseases in England and Wales, 1935–1995 (The Stationery Office Annual Abstract of Statistics)

A glossary of some of the diseases in the table

Scarlet fever Infectious disease. High fever, sore throat. Caused by streptococcal bacterial infection.

Whooping cough Infectious disease of mucous membrane of air passages. Frequent attacks of convulsive coughing caused by bacteria. Vaccination possible.

Diphtheria Infectious disease causing growth of membrane, often on tonsils, making swallowing difficult. The bacterium produces a toxin. Vaccination began in 1940.

Measles Viral infection. Causes fever and rash. Vaccine available.

Typhoid A fever with ulceration of skin and bowels. Bacteria carried in sewage which is main cause of contagion.

Tuberculosis Disease of lungs and other organs caused by bacteria. Improved hygiene, vaccination and chest X-rays have helped to minimise its effects.

Syphilis A sexually transmitted (or inherited) disease. Slow to manifest itself. Caused by a spirochaete. Results in death unless checked.

1948 • Pernicious anaemia shown to be caused by **Vitamin B12 deficiency**
• Antibiotics **chloramphenicol** and **chlortetracycline** (the first tetracycline) discovered
1949 • **Cortisone** shown to be active in rheumatoid arthritis
1950
1951
1952
1953 • ***DNA double helix*** *discovered by Watson and Crick*
1954 • *First successful* ***kidney transplantation*** *in US*
1955 • *First* ***oral treatment*** *for diabetes introduced (Germany)*
1956
1957 • ***Interferon*** *first isolated and named by Isaacs and Lindenman at NIMR, London*
• **Imipramine** shown to be effective in depression
• **Halothane** (ICI/Zeneca) anaesthetic gas introduced
1958
1959 • First **semi-synthetic penicillin** marketed
1960 • **Methicillin** launched – active against many resistant bacteria
• **Metronidazole** (Rhône-Poulenc) for parasitic and anaerobic bacterial infections
1961 • **Allopurinol** (Wellcome) developed for gout and arthritis
1962 • 1961–3 first **benzodiazepines** (Roche) for depression
• First oral contraceptive launched – **Anovlar** (Schering Health Care)
• **Azathioprine** (Wellcome) patented for immuno-suppression
1963 • **Ampicillin** (Beecham) major antibiotic discovered
1964 • **Ibuprofen/Flurbiprofen** (Boots) for arthritis and inflammation
1965 • **Propranolol** (ICI/Zeneca) a beta-blocker for heart disease
1966
1967 • **Becotide** (Allen & Hanbury's) for asthma
• *First* ***heart transplant*** *by Christiaan Barnard in South Africa*
1968 • **Sodium cromoglycate** (Fisons) breakthrough in asthma
1969 • **Salbutamol** (Glaxo) introduced for asthma
1970 • **Levodopa (L-dopa)** a major advance in Parkinson's
1971 • *Mechanism of action of* ***aspirin*** *discovered by Sir John Vane*
1972
1973 • **Tamoxifen** (ICI/Zeneca) introduced for hormone-dependent tumours
1974
1975 • ***Monoclonal antibodies*** *discovered by Kohler and Milstein (UK)*
• **Clozapine** (Novartis) first atypical neuroleptic for schizophrenia enters clinical trial
• **Clotrimazole** (Bayer) a major advance in treating fungal infections
• **Nifedipine** (Bayer) for angina and hypertension
1976 • **Atenolol** (ICI/Zeneca) a beta-blocker introduced for various heart conditions
• **Cyclosporin** (Novartis) a major advance in transplantation
• **Cimetidine** (SmithKline Beecham) launched for peptic ulcers

1977
1978 • *Birth of Louise Brown after Steptoe and Edwards develop* ***in vitro Fertilisation***
• **Ranitidine** (Glaxo) anti-ulcer treatment discovered
1979 • ***Smallpox*** *eradicated from the world*
• ***Interferon gene*** *first cloned*
1980
1981 • **Captopril** (Bristol-Myers Squibb) first ACE inhibitor for high blood pressure
1982 • **Fluconazole** (Pfizer) – key advance in treating fungal infections
1983 • *Sir John Vane awarded Nobel Prize for work on* ***aspirin*** *and* ***prostaglandins***
• *Isolation of* ***HIV*** *as the cause of AIDS*
• **Sumatriptan** (Glaxo) – major advance in migraine
1984 • **Cholestyramine** – trial shows lowering of cholestrol and coronary heart disease
• Antibiotic **Augmentin** (Beecham) launched
1985 • **Acyclovir** (Wellcome) treatment for herpes launched
1986 • ACE inhibitor **enalapril** (MSD) launched for high blood pressure
• **Orthoclone** (Ortho) for transplantation – first licensed human monoclonal antibody
1987 • **Zidovudine** (Wellcome) – first AIDS treatment launched
1988 • **Lisinopril** (ICI/Zeneca) ACE inhibitor for hypertension and heart failure
• *Nobel Prize awarded to* ***Sir James Black*** *for medicines discovery*
• **Diclofenac** (Novartis), an anti-inflammatory agent, launched
• **Erythropoietin** (Janssen-Cilag), natural red blood cell stimulator, launched in the UK
1989 • **Omeprazole** (Astra) launched for gastric ulcers
• **Simvastatin** (MSD) launched for lowering blood lipids
• **Fluoxetine** (Eli Lilly) launched for depression
1990 • ***COX-2****, a major new target for anti-inflammatory drugs, discovered by scientists at Searle*
• *First* ***gene therapy*** *experiment in a person with adenosine deaminase deficiency*
1991 • **Filgrastim** (Amgen) white blood-cell stimulant, G-CSF launched in UK
1992 • **Etidronate** (Procter & Gamble). First bis-phosphonate in UK for osteoporosis
1993
1994
1995 • **Lamotrigine** (Wellcome) – major advance launched as monotherapy in epilepsy treatment
• **Interferon beta-1b** (Schering Health Care) – first treatment for multiple sclerosis
1996 • **Olanzapine** (Eli Lilly) introduced for schizophrenia
• **Ropinirole** (SmithKline Beecham) launched for Parkinson's
• **Saquinavir** (Roche) launched – first protease inhibitor for AIDS in UK
1997 • First medicines for Alzheimer's disease available – **Donepezil** (Pfizer) and **Tacrine** (Parke-Davis)
• **Latanoprost** (Pharmacia & Upjohn) first prostaglandin analogue for glaucoma
• **Reboxetine** (Pharmacia & Upjohn) first noradrenaline reuptake inhibitor for depression

Figure 1 Time line of some major discoveries in medicines research 1948–1997

3: The British pharmaceutical industry

Search the Internet and look at the data provided (which come from The Association of the British Pharmaceutical Industry).

Here are some of the questions you may wish to address in your research and presentation.

- How important is the pharmaceutical industry in national terms to the economy?
- How has the industry grown over the years?
- Into which categories can the products of the industry be usefully divided?
- How important is the British pharmaceutical industry internationally?

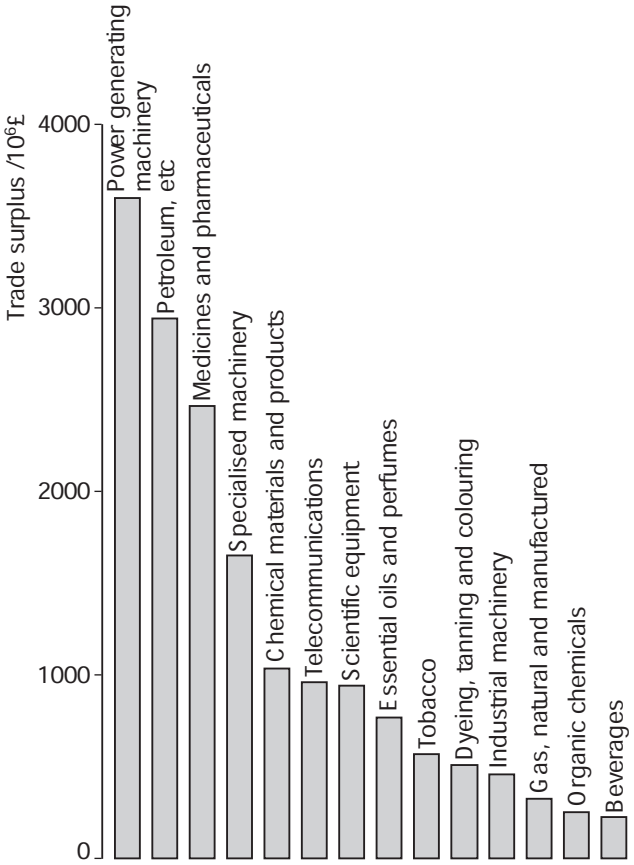
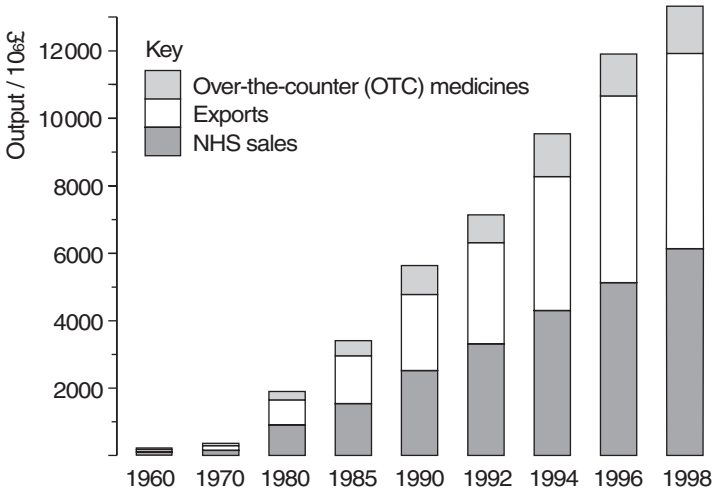


Figure 1 UK visible trade surpluses in 1998



Source: *Pharma, Facts and Figures*, Association of the British Pharmaceutical Industry, 2000

Figure 2 British pharmaceutical industry output by market sectors

	Exports /10^6£	Imports /10^6£	Balance /10^6£	Exports per employee/£	All UK exports per employee /£
1981	852	298	554	12 206	6345
1982	978	375	603	14 257	7088
1983	1074	470	604	15 817	7978
1984	1222	542	680	18 077	9420
1985	1426	590	835	21 315	10 520
1986	1533	679	853	23 991	11 205
1987	1621	786	835	24 339	12 712
1988	1735	876	859	25 628	13 675
1989	2016	1062	955	28 315	15 816
1990	2258	1158	1100	31 759	17 873
1991	2556	1371	1184	35 104	20 019
1992	2993	1663	1330	40 553	21 744
1993	3710	2019	1691	53 927	25 113
1994	4005	2304	1701	57 705	28 582
1995	4939	2812	2126	79 784	31 925
1996	5386	3107	2279	91 597	34 386
1997	5455	3192	2262	90 912*	35 496
1998	5860	3418	2442	97 667*	

Table 2 British pharmaceutical trade figures

*Subject to revision

Source: *Pharma, Facts and Figures*, Association of the British Pharmaceutical Industry, 2000

	Exports /10^6£	Imports /10^6£	Trade balance /10^6£
Germany	7741	4179	3562
Switzerland	4904	2115	2790
UK	5860	3418	2442
Ireland	2620	486	2134
France	5193	3507	1685
Sweden	2142	739	1403
Belgium	3056	2380	676
Denmark	620	471	150
Netherlands	1967	2043	–76
Austria	809	1058	–249
Italy	2344	2596	–252
Finland	138	394	–255
Portugal	87	441	–358
Greece	70	600	–530
Australia	393	988	–596
Spain	743	1572	–828
Canada	675	1525	–850
US	4817	5687	–870
Japan	686	1948	–1262

Table 3 World pharmaceutical trade figures, 1998

Source: *Pharma, Facts and Figures*, Association of the British Pharmaceutical Industry, 2000

WM2

Extraction of salicylic acid

In this activity you can extract some salicylic acid, a pharmacologically active compound from a natural source, oil of wintergreen, which is obtained in turn from the shrub* Gaultheriae procumbus*. You then learn the technique of thin-layer chromatography and use it to compare the extract with a pure sample of salicylic acid.

Requirements

- oil of wintergreen (2 cm^3)
- apparatus for heating under reflux (see Figure 1)
- sodium hydroxide solution, 2 mol dm^{-3} (25 cm^3)
- measuring cylinder (25 cm^3)
- anti-bumping granules
- small Bunsen burner or electric heating mantle
- 100 cm^3 beakers (2)
- ice (if available)
- concentrated hydrochloric acid (a few cm^3)
- Universal Indicator paper
- glass rod
- Buchner funnel and apparatus for vacuum filtration
- watch glass
- ethanol (a few cm^3)
- t.l.c. plates (silica-coated)
- small beaker to hold t.l.c. plate
- cover for the beaker
- solution of salicylic acid in ethanol (1 cm^3)
- solvent for chromatography – cyclohexane, ethyl ethanoate, ethanoic acid (200:100:1)
- dropping tubes or melting-point tubes (2)
- u.v. light source
- iodine crystals
- aluminium foil or clingfilm

CARE Ultraviolet radiation is harmful to the eyes. Do not look directly at the lamp. Follow the recommended precautions concerning eye protection.


ULTRAVIOLET RADIATION

CARE Eye protection must be worn.


WEAR EYE PROTECTION

cyclohexane


HIGHLY FLAMMABLE

glacial ethanoic acid


CORROSIVE


FLAMMABLE

ethanol


HIGHLY FLAMMABLE

ethyl ethanoate


HIGHLY FLAMMABLE

concentrated hydrochloric acid


CORROSIVE

2-hydroxybenzoic acid (salicylic acid)


IRRITANT

iodine


HARMFUL

methyl 2-hydroxybenzoate, methyl salicylate (oil of wintergreen)


HARMFUL

sodium hydroxide solution


CORROSIVE

Introduction

The ester methyl salicylate found in nature can be converted into salicylic acid. It is obtained as oil of wintergreen which used to be made by dry distillation of the leaves of the evergreen shrub *Gaultheriae procumbus* prevalent in the US. You may be familiar with the smell of the ester for it is present in many medicinal preparations (for example 'Deep Heat' and other similar ointments used to relieve muscle pain).

In this experiment you will obtain salicylic acid from the methyl ester by hydrolysis, using dilute sodium hydroxide solution.

What you do

1 Put about 2 cm^3 of oil of wintergreen (**CARE** Harmful) into a 50 cm^3 pear-shaped flask and add 25 cm^3 of 2 mol dm^{-3} sodium hydroxide solution (**CARE** Corrosive) together with a few anti-bumping granules.

2 Attach a water condenser to the flask in a vertical position (see Figure 1) and gently heat the mixture under reflux for 30 minutes. Make sure the condenser water supply is on. You can heat the flask with a small Bunsen flame below a wire gauze or use an electric heating mantle. When the mixture is boiling, condensed droplets should be falling back into the flask at a rate of about 1 drop per second.

3 Allow the mixture to cool and pour it into a 100 cm^3 beaker surrounded by cold water (and ice, if available). Using a dropper, add concentrated hydrochloric acid drop by drop (**CARE** Corrosive) until the mixture is acidic. Test the mixture, as you add the acid, with Universal Indicator paper, using a glass rod and small drops of the mixture.

4 Using a Buchner funnel, filter the product using vacuum filtration. Wash the solid with a little cold water and transfer it to a watch glass.

5 Take a few crystals of the product and dissolve them in a minimum of ethanol.

6 Take a pre-dried thin-layer chromatography plate which will fit into a small beaker (see Figure 2). About 1 cm from the bottom of the plate draw a fine pencil baseline. On the line place a small spot of the extract from stage **5**. On the baseline also put a small spot of a solution of salicylic acid in ethanol. The spots are best made using a very fine dropping pipette or a drawn-out melting-point tube. Apply a small quantity of the solution at a time; let it dry, and then add more. Try not to let the diameter of the drops exceed 5 mm.

7 Place some solvent for the chromatograpy in the beaker to a depth of about 5 mm. (**CARE** Highly flammable. Avoid breathing the vapour.)

8 Place the chromatography plate in the beaker, making sure the solvent is below the pencil line on the plate.

9 Cover the beaker, for example with aluminium foil or a watch glass. Leave the solvent to rise up. This will take about 15–25 minutes.

10 When the solvent has nearly reached the top of the plate, take the chromatogram out of the beaker. (**CARE** Avoid breathing the solvent vapour.) Place the plate in a fume cupboard and allow the solvent to evaporate.

11 You can locate the positions of the substances on the plate by examining it under u.v. light. (**CARE** Do not look directly at the light source.) View the plate by reflected light.

12 Alternatively, place the sheet in another beaker with a few crystals of iodine (**CARE** Harmful. Avoid skin contact. Use in a fume cupboard.) Cover the beaker with aluminium foil or clingfilm. The spots which were probably just visible before should show up more clearly now. After the experiment, dispose of the solvent as directed by your teacher.

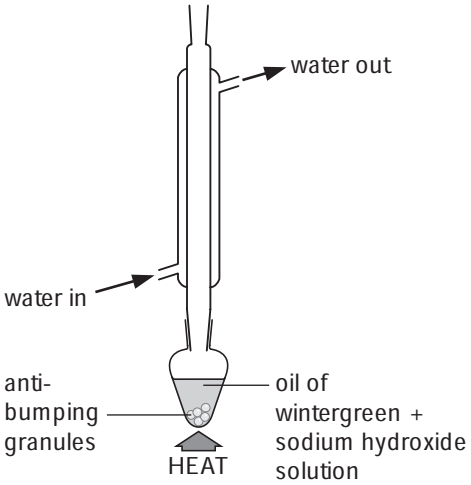


Figure 1 Apparatus for heating under reflux

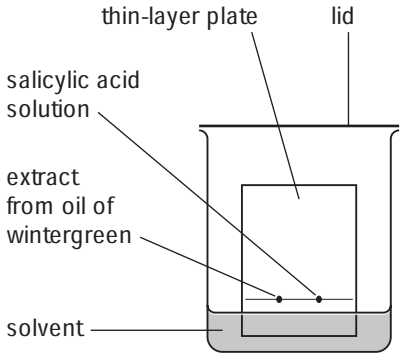


Figure 2 Apparatus for thin-layer chromatography

QUESTIONS

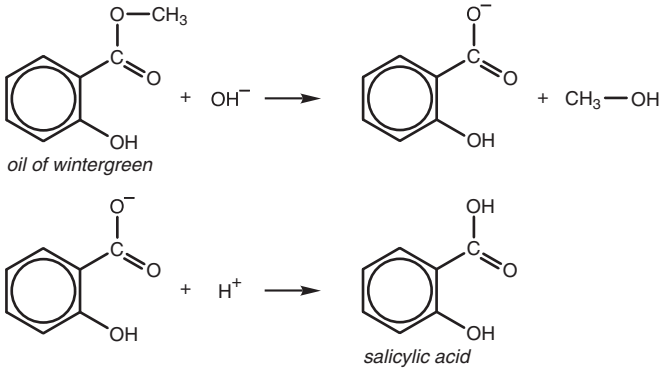
a Explain what is meant by *heating under reflux*. Why is this often necessary when heating organic liquids?

b Why is it necessary to heat the flask for such a long time?

c What do the results from the thin-layer chromatography separation tell you about the composition of the extract from oil of wintergreen?

d The equations for the reactions occurring in the experiment are shown on the right. How could you have obtained the methanol formed during the experiment if you had wished to use it?

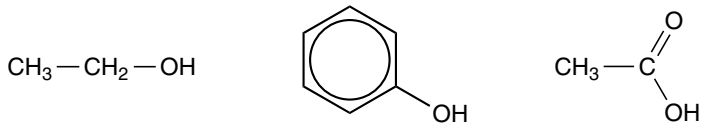
e Suggest why the mixture formed by the hydrolysis of the ester was kept as cool as possible.



WM3

Investigating the chemistry of the –OH group in various environments

The hydroxyl (–OH) group occurs in three types of organic compound: alcohols, phenols and carboxylic acids. Examples are:



ethanol *phenol* *ethanoic acid*

The object of Part 1 of this activity is to investigate the behaviour of the –OH group in these three compounds and to compare this with the behaviour of 2-hydroxybenzoic acid (salicylic acid).

In Part 2 you can compare the behaviour of the –OH group in different alcohols.

Requirements

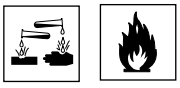
- small quantities of:
  - ethanol, labelled A
  - phenol (saturated solution in water), labelled B
  - ethanoic acid (2 mol dm^{-3} solution), labelled C
  - glacial ethanoic acid (for test **6**)
  - 2-hydroxybenzoic acid (salicylic acid), solid, labelled D
- protective gloves
- Universal Indicator solution
- iron(III) chloride solid
- potassium dichromate(VI) solution, 0.1 mol dm^{-3} (2 cm^3)
- methanol (5 cm^3)
- concentrated sulphuric acid (access to bottle)
- sulphuric acid, 2 mol dm^{-3} (5 cm^3)
- sodium carbonate solution, approximately 0.5 mol dm^{-3} (200 cm^3)
- test-tubes
- 100 cm^3 beakers (8)
- a range of alcohols:
  - propan-1-ol (3 drops)
  - butan-1-ol (3 drops)
  - butan-2-ol (3 drops)
  - 2-methylpropan-2-ol (3 drops)
- source of hot water

CARE Phenol can cause sores and blistering if spilt on the skin. Glycerol, propane-1,2,3-triol, can be applied to counteract phenol burns.

CARE Alcohols are highly flammable liquids.
Keep bottles stoppered when not in use and well away from naked flames.
Avoid skin contact and do not breathe the vapour.

CARE Dichromates(VI) irritate the skin. They are also suspected carcinogens. Avoid all skin contact. Any spillage should be washed off at once. Wear protective gloves.

glacial ethanoic acid



ethanol

2-hydroxybenzoic acid (salicylic acid)

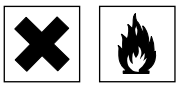
iron (III) chloride

methanol

phenol

potassium dichromate(VI) solution

propan-1-ol
butan-1-ol
butan-2-ol
2-methylpropan-2-ol

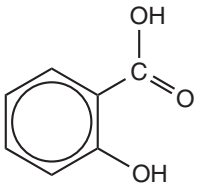


sulphuric acid

CARE Eye protection and gloves must be worn.

Part 1: Comparing ethanol, phenol, ethanoic acid and 2-hydroxybenzoic acid

- Substance A is the alcohol ethanol (**CARE** Highly flammable).
- Substance B is a solution of phenol (**CARE** Toxic and corrosive. Avoid all skin contact. Wear gloves).
- Substance C is a solution of ethanoic acid. Ask for a more concentrated sample for test **6**. (**CARE** Concentrated ethanoic acid is corrosive and has an irritant vapour. Avoid skin contact and inhalation.)
- Substance D is 2-hydroxybenzoic acid (salicylic acid) (**CARE** Irritant). The tests on D can be carried out either with the solid or with a concentrated solution, whichever seems more appropriate.



2-hydroxybenzoic acid (salicylic acid)

Look quickly through the tests you will be doing, and draw up a table to present your results.

For each test use 1 cm depth of the sample solution or a small spatula measure of solid.

What you do

1 To each of the substances in turn add a few drops of Universal Indicator solution, and record the pH value.

2 To each of the substances in turn add an equal volume of aqueous sodium carbonate and warm. Test for any gas evolved.

3 Dissolve a small spatula measure of solid iron(III) chloride (**CARE** Irritant; stains skin and clothing) in about half a test-tube of water. Divide the solution equally among four test-tubes and add a small quantity of each substance in turn to the tubes. Record any colour changes.

4 To about 1 cm depth of 0.1 mol dm^{-3} potassium dichromate(VI) solution (**CARE** Toxic) in a test-tube add 2 mol dm^{-3} sulphuric acid (**CARE** Corrosive) until the tube is half-full. Divide this solution among four test-tubes and add small quantities of each substance in turn. Warm the mixtures in their test-tubes in separate 100 cm^3 beakers of boiling water and record any colour changes. (You will need the beakers of hot water again in stage **6.**)

5 **Cautiously** smell samples of A, B, C and D in turn. Make a note of their smells.

6 Put 1 cm^3 of each substance in turn into a test-tube. In the case of ethanoic acid, use the concentrated ('glacial') acid rather than an aqueous solution. (**CARE** Concentrated ethanoic acid is corrosive and flammable.) Then to each add an equal volume of methanol and a few drops of concentrated sulphuric acid (**CARE** Corrosive). Warm each tube and its contents gently for a few minutes in hot water in a beaker. Pour the hot solutions into beakers of dilute sodium carbonate solution. This neutralises any acid which remains and so removes its smell. **Cautiously** smell the contents of the beakers. Note any changes of smell.

What do you do if the organic material catches fire?

Do not panic!

Place the test-tube in the rack and place a wet cloth over the top.

Do not run around with the test-tube on fire.

Do not use a fire extinguisher for a small fire in a test-tube.

QUESTIONS

a Look at the results of the tests for substance D. In what way does substance D show similarities to those for the other compounds?

b What conclusion can you draw about the environments of the –OH groups in 2-hydroxybenzoic acid?

Part 2: Comparing different alcohols

1 Place about 1 cm depth of 0.1 mol dm^{-3} potassium dichromate(VI) solution in a test-tube. Add 2 mol dm^{-3} sulphuric acid until the tube is half full. Then divide this mixture as equally as possible between five test-tubes.

You are going to investigate the effect of this oxidising mixture on a range of alcohols, for example ethanol, propan-1-ol, butan-1-ol, butan-2-ol and 2-methylpropan-2-ol.

2 Add 3 drops of one of the alcohols to the oxidising mixture in one of the tubes. Be careful not to add too much alcohol. (**CARE** Alcohols are highly flammable. Keep the bottle well away from naked flames.)

Carefully **warm** the contents of the tube until they just begin to boil. (**CARE** Do not continue to boil the liquid in case alcohol vapour catches fire.)

3 Label the tube and leave it to stand. Repeat the procedure in step **2** with each of the other alcohols.

4 Make a note of any changes of appearance of the mixtures in the tubes. Work out what has happened in each case, and present your results in the form of a table showing the structural formulae of the alcohols and any products which are formed.

QUESTIONS

c There is a pattern in the behaviour of the alcohols towards oxidation. Describe the pattern.

d There are three different structural types of alcohol in this investigation. Suggest why the three types behave differently. Building models of a series such as butan-1-ol, butan-2-ol and 2-methylpropan-2-ol might help you see more clearly what is happening.

WM4

Interpretation of the mass spectrum of salicylic acid

In this activity you will practise some of the ideas you are learning about mass spectrometry, and see how chemists can identify a substance from its mass spectrum.

***Before starting the activity you should be familiar with the technique of mass spectrometry. If necessary, read about this in* Chemical Ideas 6.5.**

This activity guides you through a sequence of steps which a chemist could take when using mass spectra data to confirm the structure of a substance extracted from willow bark and given the name salicylic acid. You can record the evidence as you acquire it on the **Information Sheet** (*Mass spectrum data*). Remember research chemists, when publishing their findings, must supply as much evidence as possible to confirm the structure they propose for a compound even if, when more limited evidence is available, they already feel confident about the structure.

Finding possible formulae

1 Look first at Figure 1, the mass spectrum of salicylic acid. Measure the relative abundance of the six marked peaks. Record the mass and abundance of each peak in the appropriate box towards the bottom of the **Information Sheet**.

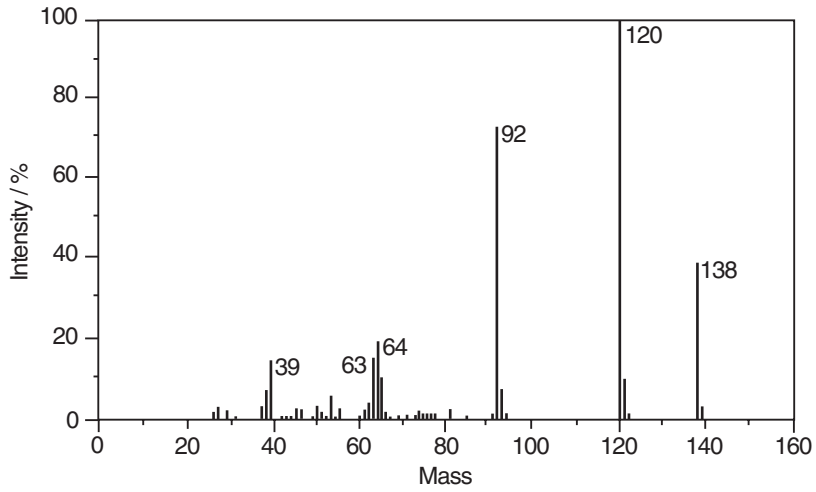


Figure 1 The mass spectrum of salicylic acid

2 Decide which of these peaks gives the molecular mass of the compound.

A computer database can list the possible molecular formulae that will fit this value. There can of course be a very large number. One limited database gives 38 compounds for this molecular mass. Add this number to the **Information Sheet**.

Using isotope peaks to establish the formula

The number of possible compounds can be reduced by using the high-resolution mass spectrum facility. For the willow bark extract this gives an accurate molecular mass of 138.0317. In the same database there are only two compounds with this value: they are $C_2H_7ON_4Cl$ and $C_7H_6O_3$. Add these possible molecular formulae to the **Information Sheet**.

Naturally occurring carbon consists of a mixture of isotopes. Most carbon is ^{12}C, but ^{13}C is present as 1.1% of the sample. If the active chemical in the extract contains one carbon atom in its molecule, the molecular ion peak at a mass M should be accompanied by another peak at M+1 with approximately 1.1% the intensity of the molecular ion peak. Two carbons will give rise to an M+1 peak of approximately 2.2% relative intensity, and so on.

This means that for one carbon atom, the ratio of the intensities of M : M+1 peaks will be 98.9 : 1.1; for two carbon atoms the ratio will be 97.8 : 2.2, and so on.

3 Figure 2 gives a more accurate mass spectrum for salicylic acid. This spectrum shows any isotope peaks which exist. The intensities of the molecular ion peaks have been accurately measured and are given on the **Information Sheet**.

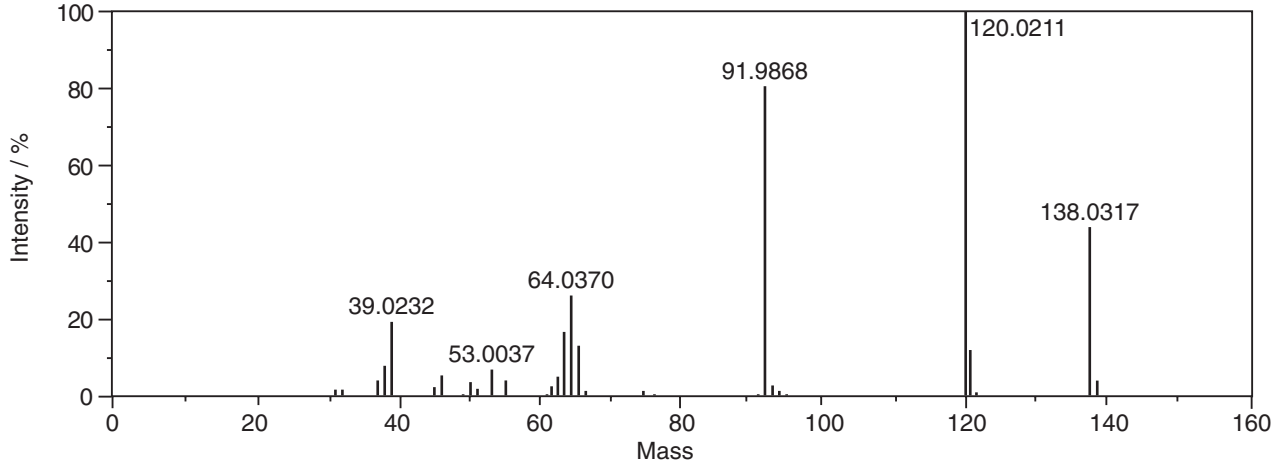


Figure 2 A more accurate version of the mass spectrum of salycylic acid

Using a database to establish which isomer it is

A web site, which supplies mass spectra of compounds, indicates that there are three isomers with the molecular formula $C_7H_6O_3$. The mass spectra of these compounds provides the following information.

2-hydroxybenzoic acid						
Peak	120	92	138	64	39	63
Intensity	100	73	39	19	14	14
3-hydroxybenzoic acid						
Peak	138	121	93	65	39	91
Intensity	100	75	23	16	12	12
4-hydroxybenzoic acid						
Peak	121	138	65	93	39	63
Intensity	100	83	24	22	18	10

Table 1 Mass spectra of the three hydroxybenzoic acids

4 Which of these matches the spectrum of salicylic acid? Write the chemical name and structure of salicylic acid on the **Information Sheet**.

Confirming the structure using the fragmentation pattern

Mass spectrometrists can find out a lot about the structure of a molecule by looking at the fragmentation pattern in the mass spectrum. This provides an additional way of distinguishing 2-hydroxybenzoic acid from its two isomers, 3- and 4-hydroxybenzoic acid. It may help with this activity to build models of the three molecules.

5 Using Table 1, write down the peaks in the mass spectrum of 2-hydroxybenzoic acid that are not shared by 3- and 4-hydroxybenzoic acid.

6 Which well-known molecule with $M_r = 18$ might be lost in the conversion:

138 (the whole molecule) → 120

in 2-hydroxybenzoic acid?

7 What are the two most likely ways in which the molecule in **6** can arise from 2-hydroxybenzoic acid? Show this by ringing the relevant atoms on drawings of the structure of the molecular ion of 2-hydroxybenzoic acid (Figure 3).

8 3- and 4-hydroxybenzoic acid do not have the same fragmentation pattern because they cannot form an ion of mass = 120. Explain why this ion cannot form.

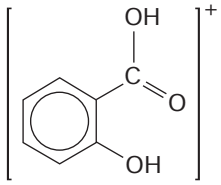


Figure 3 The molecular ion of 2-hydroxybenzoic acid

Information Sheet: Mass spectrum data

Approximate molecular mass
(found by fast, low-resolution scan)

Number of possible molecular formulae

Accurate molecular mass
(measured by slower, high-resolution scan)

Possible molecular formulae

Isotope peaks

A group of peaks starting at the parent ion peak, due to isotopes of carbon. These peaks may be small on the spectrum, but are given here scaled up so that the largest peak in this group has abundance 100.

Molecular ion peak = 100% Indicated number of carbon atoms:
M + 1 peak = 8.2%

Six largest peaks in the spectrum

Mass						
Relative abundance						

Identity of this sample

WM5.1

A preparation of aspirin

This activity is an example of an organic preparation. You will be able to purify and test your product.

Requirements

- 100 cm^3 conical flask
- 10 cm^3 measuring cylinders (2)
- 100 cm^3 beaker
- glass rod
- apparatus for vacuum filtration
- Hirsch funnel
- 2-hydroxybenzoic acid (salicylic acid) (2 g)
- ethanoic anhydride (4 cm^3)
- concentrated sulphuric acid (5 drops)
- ethanoic acid (glacial) (4 cm^3)
- water bath containing crushed ice
- source of hot water
- test-tubes (4)
- aspirin (1 crystal)
- neutral iron(III) chloride solution (1 cm^3)

ethanoic acid

CORROSIVE

ethanoic anhydride

CORROSIVE

2-hydroxybenzoic acid (salicylic acid)

IRRITANT

iron (III) chloride

IRRITANT

concentrated sulphuric acid

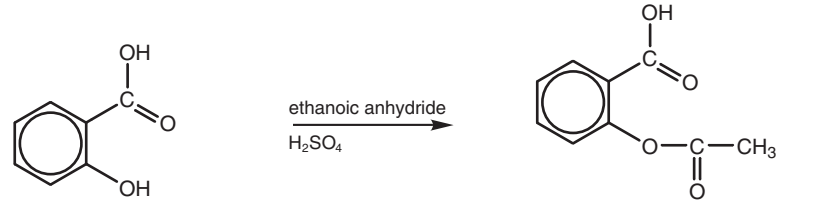
CORROSIVE

CARE Eye protection must be worn.

WEAR EYE PROTECTION

Introduction

Aspirin (acetylsalicylic acid or 2-ethanoylhydroxybenzoic acid) can be made by a variety of methods but they all start from 2-hydroxybenzoic acid (salicylic acid).



2-hydroxybenzoic acid *2-ethanoylhydroxybenzoic acid*

What you do

1. Shake 2 g of 2-hydroxybenzoic acid (salicylic acid) (**CARE** Irritant) with 4 cm^3 of ethanoic anhydride (**CARE** Corrosive) in a 100 cm^3 conical flask.
2. Add five drops of concentrated sulphuric acid (**CARE** Corrosive) and continue agitating the flask for about 10 minutes. Crystals of aspirin will appear and soon the whole will form a crystalline mush.
3. Dilute by stirring in 4 cm^3 of cold glacial ethanoic acid (**CARE** Corrosive) and cool by placing in a water bath containing crushed ice.
4. Filter off the crystals using a Hirsch funnel (a small funnel for vacuum filtration), washing once with ice cold water. Reserve a few crystals for testing later.
5. Place the crude aspirin in a 100 cm^3 beaker. Add hot, but not boiling, water until it dissolves. Cool and filter off the crystals. This process is known as **recrystallisation** and is a way of purifying a solid product.
6. Take four test-tubes and add 2 cm^3 of distilled water to each.
   To one tube add one crystal of the product before recrystallisation and shake.
   To another add one crystal of the recrystallised product and shake.
   To another add one crystal of 2-hydroxybenzoic acid and shake.
   To the last add one crystal of known pure aspirin and shake.
   To each tube in turn add 2 drops of neutral iron(III) chloride solution (**CARE** Irritant) and shake.

QUESTIONS

a On the basis of your observations in part **6**, was your product pure aspirin?

b A student obtained 1.7 g of dry product. Calculate the percentage yield.

c Explain why recrystallisation should produce a purer product.

d There are several ways of testing the purity of the product:

i neutral iron(III) chloride solution

ii thin-layer chromatography

iii melting point.

Explain with the aid of a diagram how you would use thin-layer chromatography to test the purity of a sample of aspirin.

WM5.2

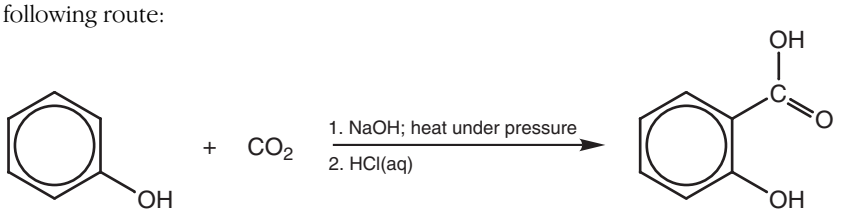
Using spectroscopy

In this activity you will analyse the infrared and mass spectra of some organic molecules used in the synthesis of the medicine, aspirin.

For this activity you will need to refer to the characteristic i.r. absorption frequencies given in the **Data Sheets**.

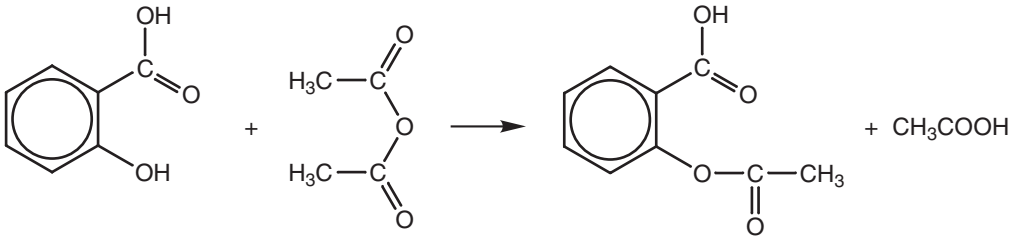
Aspirin, 2-ethanoylhydroxybenzoic acid, is manufactured commercially from phenol by the following route:

Step 1



phenol (A) — *2-hydroxybenzoic acid* (B)

Step 2



ethanoic anhydride — *aspirin* (C)

QUESTIONS

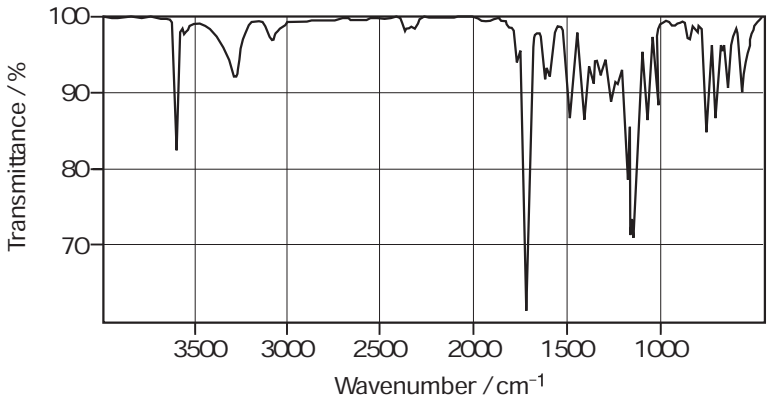
a The i.r. and mass spectra of compounds A, B and C are given in Figures 1–3 on the next sheet.
Identify, with as much explanation as possible, which set of spectra (Figure 1, 2 or 3) corresponds to each of the compounds A, B and C.

b Use the data of % composition by mass, given in Table 1, to confirm your answer to part **a**.

Compound	%C	%H	%O
Figure 1	60.9	4.3	34.8
Figure 2	60.0	4.4	35.6
Figure 3	76.6	6.4	17.0

Table 1 Composition by mass

Figure 1



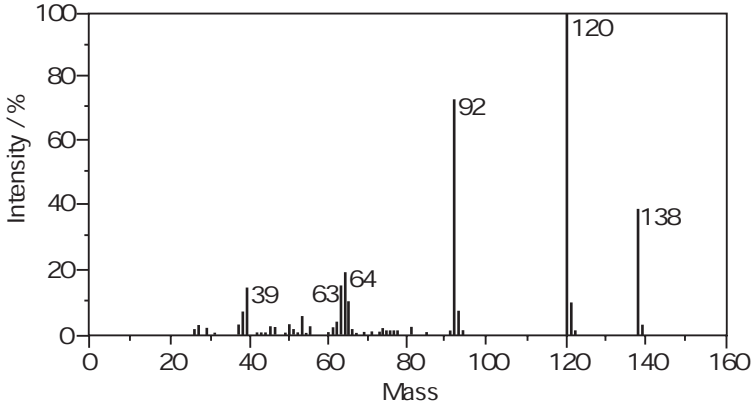
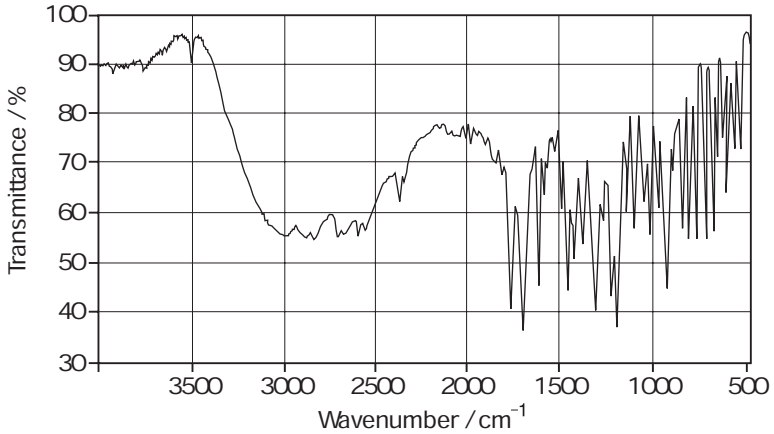


Figure 2



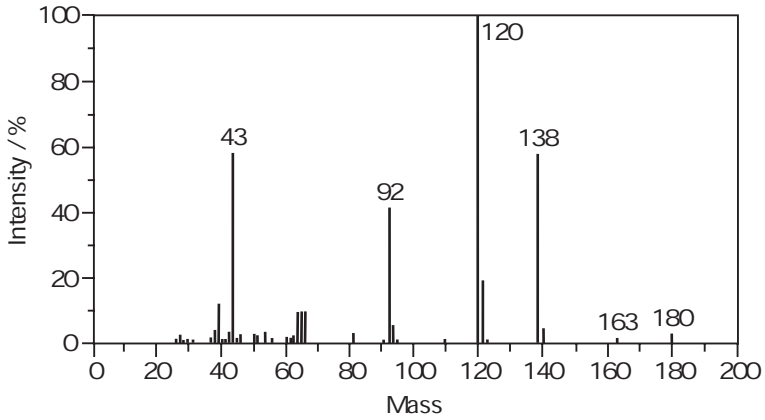
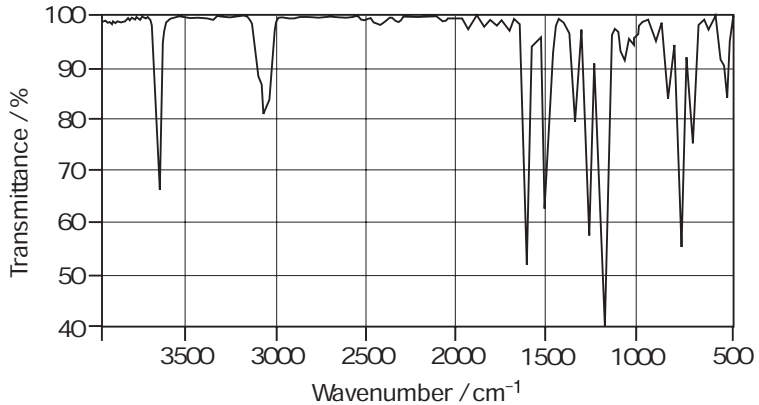
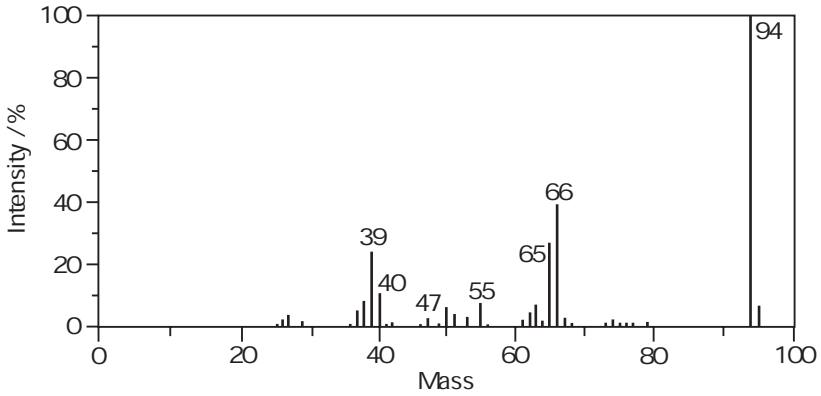


Figure 3





WM6

An aspirin assay

In this activity you will follow the method used by hospital analysts to check the purity of aspirin samples before they are considered fit to be used on the wards.

Requirements

- aspirin tablets (300 mg)
- mortar and pestle
- specimen tubes (3)
- access to a balance
- 100 cm^3 conical flask
- 95% ethanol (30 cm^3)
- 10 cm^3 measuring cylinder
- sodium hydroxide solution, 0.1 mol dm^{-3} (60 cm^3)
- burette
- Phenolphthalein indicator

ethanol

HIGHLY FLAMMABLE

CARE Eye protection must be worn.

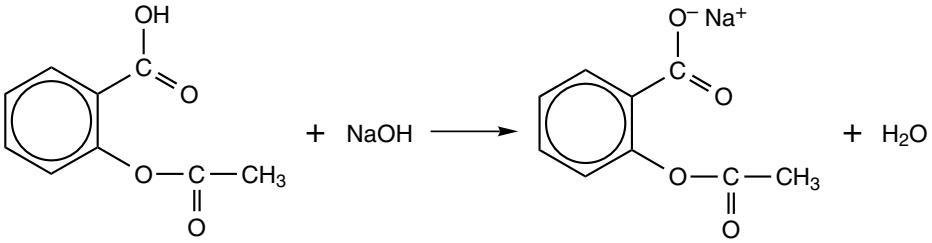
WEAR EYE PROTECTION

What you do

1 Grind up 1 aspirin tablet using a mortar and pestle.
2 Transfer as much of the powder as possible into a specimen tube. Weigh the tube to an accuracy of 1 mg, and record the mass.
3 Use a measuring cylinder to place 10 cm^3 of 95% ethanol into a 100 cm^3 conical flask. Add a few drops of Phenolphthalein indicator. Then add as much of the powdered aspirin as you can from the specimen tube.
4 Reweigh the specimen tube to the same accuracy as before and record the mass.
5 Swirl the conical flask carefully until all the aspirin powder has dissolved. Do not allow any of the solution to splash out of the flask.
6 Titrate the solution in the flask with 0.1 mol dm^{-3} sodium hydroxide solution from a burette (**CARE** Eye protection *must* be worn). Record the volume needed to produce the first tinge of pale pink colour in the indicator. This measures the end-point of the titration.
7 Repeat the procedure at least once more, starting with a fresh aspirin tablet.

Calculation

The equation for the reaction used in the assay is:



Notice that 1 mol of aspirin reacts with 1 mol of sodium hydroxide. (Actually, if you were to heat the reaction mixture, a further reaction would take place and the ester would be hydrolysed. But at room temperature this does not happen.)

1 mol of aspirin has mass of 180.2 g.

Concentration of sodium hydroxide solution = …… mol dm^{-3}

8 Calculate the purity of the aspirin tablet, using Table 1.

	Assay 1	Assay 2	Assay 3
Mass of specimen tube + aspirin powder/g			
Mass of specimen tube after pouring out aspirin powder/g			
Mass of aspirin used/g			
Volume of sodium hydroxide solution used/cm^3			
Amount of sodium hydroxide used/mol			
Amount of aspirin which reacted with sodium hydroxide/mol			
Mass of aspirin which reacted with sodium hydroxide/g			
Aspirin in powder used/%			

Table 1

Evaluating your results and procedures

a What is the percentage error in your measurement of the mass of aspirin used and in the volume of sodium hydroxide solution used?

b Identify any stages in your *procedure* which could have led to errors. Comment on the range of answers obtained for your three assays.

c Bearing in mind the sources of error you have discussed above, how many significant figures do you think you are justified in using for the percentage of aspirin in the powder?

9 Accurately weigh one aspirin tablet. Use your assay results to calculate the mass of aspirin contained in the tablet. Suggest why the mass of the tablet and the mass of aspirin may be different. Why do you think the tablets are described as '300 mg' tablets?

WM8

Which product should a pharmaceutical company develop?

In the exercise that follows, you are asked to take on the role of a manager in a pharmaceutical company helping to make decisions about which products your company should develop.

General information

It is a very long process from the initial discovery of a compound which has some pharmacological activity to the final marketing of a medicine. At any one time, several different compounds may be available for further development, but the process is so expensive that a company must select only a limited number of compounds for further investigation.

Policies are reviewed at regular intervals and the progress of work on any one compound will be scrutinised at several key stages of development.

In this exercise, you need to be organised into groups which represent different management groups in a company. Some of the groups will make an 'initial technological and economic appraisal' of two potential pharmaceuticals; the other groups will make a 'second technological and economic appraisal' of the same two compounds.

Initial technological and economic appraisal

This is the stage at which the compounds under review have been identified, synthetic routes have been established and some preliminary testing for activity and toxicity is complete. The research division of the company have put forward two compounds, AP1011 and H2202, to be considered for further development.

You must consider the evidence available and produce, for each of these compounds, a summary of the arguments for and against taking up the option of further development work. Since the company budget may only allow for work on one of these, you should also recommend which seems the more promising compound to develop.

Second technological and economic appraisal

This comes after further development has been carried out. For the purpose of the exercise, it is assumed that the company adopted both compounds at the earlier stage and has since completed clinical trials and further studies of the behaviour of the compound in the body. Once again, the available evidence must be examined to produce a summary of the points for and against proceeding with each of the compounds, and you must recommend which should be developed and marketed.

When the evaluations have been completed, groups which have made the initial appraisal should compare their conclusions with groups which made the second appraisal.

Initial technological and economic appraisal

Technical information on compound AP1011

This compound has antipyretic (fever-reducing) and analgesic (pain-relieving) properties and so may be suitable for conditions such as toothache, headache, neuralgia, sciatica, period pains and pains in ligaments, muscles and joints. It also reduces swelling and pain in arthritis and rheumatic fever. There is some evidence that it may inhibit blood-clotting.

Preparation and stability AP1011 may be synthesised simply in two stages, both with high yield, from readily available and cheap raw materials. It seems likely that scaling-up of this synthesis will present no major problems. Estimated cost of large-scale production is approximately 20p per 100 g. AP1011 has already been shown to be stable in dry conditions for over two years.

It must be administered by mouth and can be formulated into tablets or as powder for dispersal in water.

Action AP1011 is a drug precursor in that it is hydrolysed in the stomach and small intestine to form an active metabolite which is absorbed through the intestinal wall into the bloodstream.

Indications from preliminary trials No major side-effects have been found. There is no evidence of carcinogenic effects. The compound may produce stomach irritation and causes nausea, vomiting and/or stomach bleeding in a few cases.

Overdose can lead to a number of symptoms, including confusion and respiratory problems.

The LD_{50} is estimated to be 1750 mg/kg of body weight.*

* The toxicity of a compound is tested by finding the dose which will kill 50% of a trial group of animals (usually rats). The dose is measured in terms of the number of milligrams of compound per kilogram of body weight of the animal.

Initial technological and economic appraisal

Technical information on compound H2202

This compound has mild sedative and hypnotic (sleep-inducing) effects and could thus fill the gap between mild pain-relieving sedatives like paracetamol and the benzodiazepines like Valium and Mogadon, which can be addictive with prolonged use.

Preparation and stability Synthesis is by a two-stage process from benzene-1,2-dicarboxylic acid and glutamic acid. Both are currently manufactured as intermediates in the production of other compounds. Scaling-up is likely to be moderately complex and the predicted cost is about £7.00 per 100 g.

The compound should be taken by mouth and can be made up into tablets. It is stable in storage.

Action H2202 is hydrolysed by the body reforming benzene-1,2-dicarboxylic acid, glutamic acid and ammonia.

Indications from preliminary trials Side-effects are almost nil. H2202 shows no toxic effects (even on fairly extreme overdose), and does not affect the liver or kidneys.

Prolonged high dosage can lead to slight disturbance of the nervous system, but this is reversed on reduction of the dose.

The LD_{50} is estimated to be 5000 mg/kg of body weight.*

* The toxicity of a compound is tested by finding the dose which will kill 50% of a trial group of animals (usually rats). The dose is measured in terms of the number of milligrams of compound per kilogram of body weight of the animal.

Second technological and economic appraisal

Technical information on compound AP1011

AP1011 has both antipyretic (fever-reducing) and analgesic (pain-relieving) properties. It is recommended for conditions involving 'mild' pain, including toothache, headache, neuralgia, sciatica, period pains and pains in ligaments, muscles and joints. For these purposes the dose rate is 600–900 mg repeated every 3–4 hours to a maximum of 4 doses in any 24 hours.

It has been shown to be remarkably effective in reducing swelling and pain in arthritis and rheumatic fever. For these purposes higher doses of up to 4 g per day (with corresponding increase in side-effects) may be needed.

It also has an effect of inhibiting blood-clotting and so may be used in low dose (100–200 mg per day) in inhibiting thrombosis.

Manufacture, storage and distribution AP1011 can be prepared on the semi-technical scale (kilogram quantities) in high purity and good yield. The predicted manufacturing cost is 35p per 100 g. There are no problems with large-scale supply of feedstock chemicals of adequate purity.

The suggested presentations are as tablets or powders (which can be dispersed in water) containing 300 mg each. Both these forms and the bulk compound are stable indefinitely under dry storage conditions.

Predicted cost per normal daily dose compared with some competitor drugs (based on use as analgesic):

AP1011	1p
Panadol	6p
Paracetamol	2p

AP1011 must be administered by mouth and can be formulated into tablets or as powder for dispersal in water.

Action AP1011 is hydrolysed in the stomach and small intestine to form an active compound which is absorbed through the intestinal wall into the bloodstream.

It acts by inhibiting the synthesis of prostaglandins (PGs). These are complex cyclic carboxylic acids and are identified by letter labels (PGE, PGF, etc.). They have many functions, among which are production of redness, heat, swelling and pain. Reductions in these symptoms are therefore experienced after taking the medicine. Some aspects of blood-clotting are also affected by prostaglandins, hence AP1011 is of use in inhibiting thrombosis.

Side-effects/contra-indications AP1011 appears effective and relatively safe in use. Long-term use does not lead to tolerance* or dependence†.

AP1011 has no effect on the central nervous system, liver or kidneys except in overdose. It can produce stomach bleeding, but this is in small amount and usually unnoticed by the patient (normal persons may experience slight stomach bleeding without taking the medicine).

0.2% of the population are intolerant of the compound and up to 28% of chronic asthma sufferers may have a bout of asthma brought on by using it.

Blood disorders such as anaemia can be caused in some patients, mainly as a result of blood loss. These cases recover on removal of the medicine and where essential symptoms may be controlled using iron tablets.

If taken during early pregnancy, AP1011 may induce abortion.

Cases of prolonged labour and excess bleeding in mother and baby at birth have been attributed to the medicine.

No tumour-inducing or carcinogenic effects related to the medicine have been observed.

The LD_{50} is estimated to be 1750 mg/kg of body weight.‡

Care should be taken when prescribing to patients already taking other medicines.

Symptoms of overdose are tinnitus (ringing in the ears), dizziness, confusion and respiratory problems.

AP1011 lowers blood-clotting ability and can produce longer bleeding times after wounds, but this effect is of benefit in the prevention of thrombosis.

* 'Tolerance' refers to the ability of the body to build up resistance to a compound so that larger and larger doses are needed to produce a given effect.

† Addiction to the compound.

‡ The toxicity of a compound is tested by finding the dose which will kill 50% of a trial group of animals (usually rats). The dose is measured in terms of the number of milligrams of compound per kilogram of body weight of the animal.

Second technological and economic appraisal

Technical information on compound H2202

H2202 is a mild sedative and hypnotic (sleep-inducing) drug. It fills the gap between mild pain-relieving sedatives such as paracetamol, and benzodiazepines like Valium and Mogadon which are addictive with prolonged use. H2202 is neither addictive nor toxic in overdose.

It is especially useful as a sedative during labour but can also be given to women who complain of nervousness, inability to sleep, or morning sickness during pregnancy. It also has more general uses for mild sleep induction.

Dose rate as a sedative is 25 mg up to three times a day.

As a hypnotic, dose rate is up to a maximum of 200 mg.

Manufacture, storage and distribution A satisfactory semi-technical scale production process has been established and no difficulty is foreseen in scaling this up for full production. The process is relatively simple, but the eventual cost of manufacture is estimated as about £35 per 100 g, mainly due to the cost of using manufactured chemicals as feedstocks.

H2202 is administered by mouth in tablet form. Normal tablet-forming equipment can be used to produce tablets in two presentations, containing either 25 mg or 100 mg of the active compound each.

Both the tablets and the bulk compound are stable in dry storage.

The principal competitors (e.g. Mogadon and Valium) are addictive. H2202 is non-addictive and has a substantially higher LD_{50}.*

Action H2202 is a white crystalline solid. It is hydrolysed in the body giving benzene-1,2-dicarboxylic acid, glutamic acid and ammonia. The mode of action in the body is not yet fully understood.

Side-effects/contra-indications In many patients there are no side-effects from small doses over short periods. Some patients experience one or more of the following side-effects: giddiness, nausea, shivering, constipation or symptoms similar to a 'hangover'. In a few cases effects on circulation are reported producing coldness or 'pins-and-needles' in hands or feet. These effects clear up on removal of the medicine.

At high dosage, disturbance of the central nervous system can take place and trembling is noticeable. Dosage over a long period can lead to polyneuritis, a nervous degeneration which is non-reversible. In pregnant women, H2202 can cross the placenta (the organ in the womb which provides the developing embryo with nutrients), hence there is a possible effect if the medicine is taken during the first two months of pregnancy.

* The toxicity of a compound is tested by finding the dose which will kill 50% of a trial group of animals (usually rats). The dose is measured in terms of the number of milligrams of compound per kilogram of body weight of the animal.

Technical and economic appraisal: management group briefing and evaluation report form

Attached to this form are reports from Research Division on compounds for which they have completed preliminary screening since our last meeting. Please produce an evaluation of the information for each of these and recommend suitability for further development and clinical trials, using the points which follow.

Compound under evaluation: .....

1 Activity

a What types of activity are already established?
What symptoms/illnesses/conditions may be treatable?
Is this likely to lead to a large market for the product?

b Are there indications of any other possible types of activity?
What additional uses might result from this?
Does this lead to a large additional potential market?

2 Manufacture, storage, delivery

a Manufacture: comment on likely costs and difficulty

b Stability: will it be possible to hold stocks for a reasonable period?

c Can the medicine be formulated in an attractive and convenient way?

d How will the likely cost compare with competitor medicines?

3 Contra-indications

a Known side-effects:

b Effects of overdose:

c LD_{50} calculated for average adults (body weight 65 kg)

d Are there any groups of people who should not use the medicine?

4 Risk/benefit ratio

a Do the benefits of using the medicine outweigh the side-effects for the large majority of those likely to use the medicine? (Circle the appropriate balance)
very good / good / acceptable / poor / very poor

5 Cost/benefit ratio

a How does the cost of the treatment compare with the value of its effects?
very good / good / acceptable / poor / very poor

6 Recommendation

a Should we proceed to clinical trials of this compound?

b If so, what special points will need to be evaluated?

c Please compare the potential of this compound with any other considered at this stage, and recommend an order of preference for development funding.

WM9

Check your notes on *What's in a Medicine?*

This activity helps you get your notes in order at the end of this unit.

Use this list as the basis of a summary of the unit by collecting together the related points and arranging them in groups. Check that your notes cover the points and are organised in appropriate ways. Remember that you will be coming back to many of the ideas in later units.

Most of the points are covered in the **Chemical Ideas**, with supporting information in the **Storyline** or **Activities**. However, if the *main* source of information is the Storyline or an Activity, this is indicated.

- The difference between primary, secondary and tertiary alcohols in terms of their structures.
- Be able to recognise members of the following homologous series: phenols, aldehydes, ketones, carboxylic acids and esters.
- The use of systematic nomenclature to name carboxylic acids and esters.
- The characteristic properties of alcohols and phenols, including: acidic nature, oxidation of alcohols to carbonyl compounds and carboxylic acids with acidified dichromate(VI) solution, dehydration of alcohols to form alkenes, test with iron(III) chloride solution, and ester formation with carboxylic acids.
- The meaning of the term: *elimination reaction*.
- The increasing relative strengths as acids of alcohols, phenols and carboxylic acids.
- The technique of heating under reflux for reactions involving volatile liquids (**Activity WM2**).
- The technique of thin-layer chromatography (t.l.c.) and the interpretation of results (**Activity WM2**).
- How the following forms of spectroscopy can be used for the elucidation of molecular structure: mass spectrometry (m.s.) and infrared spectroscopy (i.r.).
- The interpretation of mass spectra (molecular ion and significance of the fragmentation pattern) for salicylic acid and simple compounds containing a limited range of functional groups (hydroxyl, carbonyl, carboxylic acid and ester groups).
- The interpretation of infrared spectra for salicylic acid and simple compounds containing a limited range of functional groups (hydroxyl, carbonyl, carboxylic acid and ester groups).
- How more effective medicines can be obtained by modifying the structure of existing medicines (**Storyline WM5**).
- The procedures used in developing and establishing the safety of a medicine (**Storyline WM8** and **Activity WM8**).