Catalyst

GCSE Science Review

Volume 14
Number 1
September 2003

Book tokens worth £5
SEE CENTRE PAGES

Discovering DNA

FREE
EXAM REVISION NOTES
WORTH £7.95
FOR EVERY SUBSCRIBER

philip allan
UPDATES
www.philipallan.co.uk

EDITORIAL TEAM

Nigel Collins
King Charles I School, Kidderminster

David Moore
St Edward's School, Oxford

David Sang
Author and editor

Jane Taylor
Sutton Coldfield Grammar School for Girls

Editorial telephone
01562 753964

ADVISORY PANEL

Eric Albone
Clifton Scientific Trust

Tessa Carrick
Founder Editor, CATALYST

David Chaundy
Founder Editor, CATALYST

Peter Finegold
The Wellcome Trust

Chantelle Jay
Biotechnology and Biological Sciences Research Council

Peter Jones
Science Features, BBC

David Knee
CREST Awards

Sarah Leonard
Science Museum

Ted Lister
Royal Society of Chemistry
Founder Editor, CATALYST

Ken Mannion
Sheffield Hallam University

Andrew Morrison
Particle Physics and Astronomy Research Council

Jill Nelson
British Association

Silvia Newton
Cheshunt School and Association for Science Education

Toru Okano
The Rikkyo School in England

John Rhymer
Bishop's Wood Environmental Education Centre

Julian Sutton
Torquay Boys' Grammar School

Nigel Thomas
The Royal Society

© PHILIP ALLAN UPDATES 2003
ISSN 0958-3629

Publishing Editor: Jane Buekett.

Artwork: Gary Kilpatrick.

Reproduction by De Montfort Repro, Leicester.

Printed by Raithby, Lawrence and Company, Leicester.

Catalyst

The front cover shows the structure of DNA (Alfred Pasieka/SPL).

Free book for every subscriber!

A single subscription to CATALYST, Volume 14, 2003/2004 is available to individuals at £15.95 per annum. Bulk orders of three or more subscriptions are available at the greatly reduced rate of **£8.95** per subscription, provided all copies can be mailed to the same addressee for internal distribution.

CATALYST is published four times through the school year, in *September*, *November*, *February* and *April*. Orders can be placed at any time during the year, and the issues already published will be supplied automatically. Only orders for complete volumes can be accepted.

Every subscriber will also receive a copy of Philip Allan Updates' *GCSE Science Exam Revision Notes* (worth £7.95) FREE with the November issue. This offer applies only to UK subscribers.

The above rates apply only for UK addresses. Overseas rates are available on request.

Enquiries

For more information or to place an order, contact CATALYST Subscriptions at Philip Allan Updates.
tel: 01869 338652 fax: 01869 337590
e-mail: sales@philipallan.co.uk www.philipallan.co.uk
Published by Philip Allan Updates, Market Place, Deddington, Oxfordshire OX15 0SE.

Whenever anyone makes a significant discovery in science, or unravels a problem, they publish their results in a scientific journal. Fifty years ago this year, the journal *Nature* contained a short note by Francis Crick and James Watson, in which they proposed a structure for DNA. Their note, which occupied only one page of the journal, is reproduced above right. For Watson and Crick everything had quite suddenly fallen into place.

> We wish to suggest a structure for the salt of deoxyribose nucleic acid (D.N.A.). This structure has novel features which are of considerable biological interest....This structure has two helical chains each coiled round the same axis.

DNA is two stranded, each strand coiled into a helix around the other one, making a **double helix**. The strands are made of alternate deoxyribose sugar (S) and phosphate groups (P), linked to each other by strong covalent bonds. Strong bonds also occur between each sugar group and an inward-pointing unit called a **base**. There are two types of bases, purines and pyrimidines. Watson and Crick recognised how these bases could pair up and how they fitted within the helix (Figure 1).

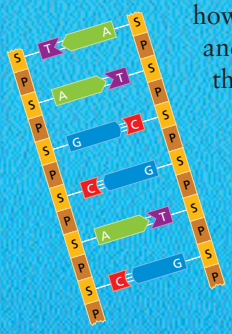
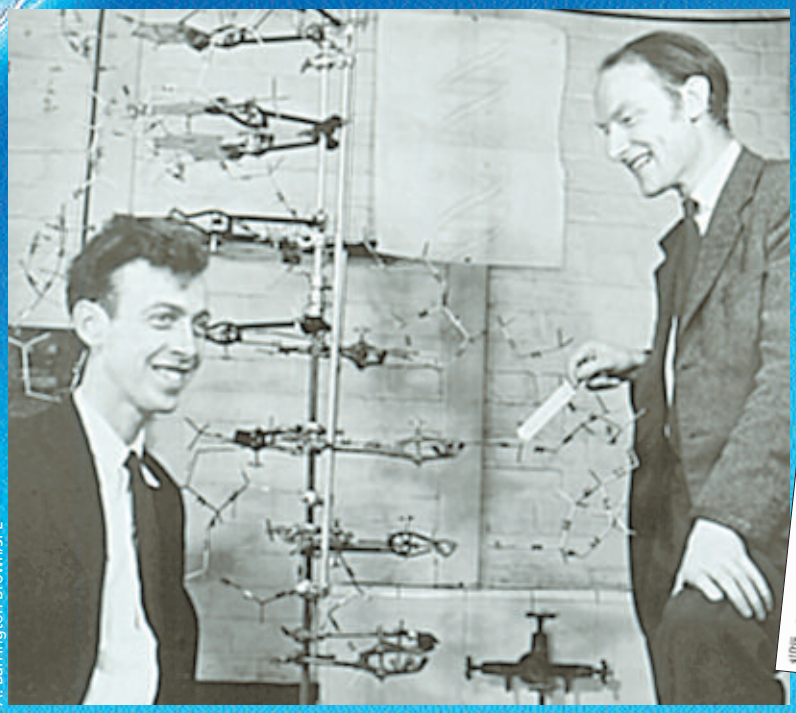


Figure 1 *The structure of DNA. These two strands are twisted into a helix (see above)*



A. Barrington Brown/SPL

Far left: *James Watson and Francis Crick stand beside their model of DNA which is now on display in the Science Museum, London. The drawing from their* Nature *paper is behind them on the wall*

Left: *A facsimile of the* Nature *paper*

Discovering DNA

The novel feature of the structure is the manner in which the two chains are held together by the purine and pyrimidine bases.

There are four bases in DNA:

A (adenine) T (thymine)
G (guanine C (cytosine).

...it is found that only specific pairs of bases can bond together. These pairs are: adenine (purine) with thymine (pyrimidine), and guanine (purine) with cytosine (pyrimidine).

The two strands are cross-linked. Relatively weak hydrogen bonds formed between the inward-pointing bases zip the two strands together. Adenine always pairs with thymine and guanine always pairs with cytosine. When DNA 'unzips', these bases are re-exposed.

COPYING THE CODE

Not only did Watson and Crick work out the structure of DNA, they also spotted something else. They knew that inherited information had to be copied exactly from cell to cell — there cannot be mistakes in the instructions for making an organism or it will not develop or work properly.

It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

DNA can copy itself by unzipping from one end. As it unzips, a new strand of DNA is built by adding bases, and sugar and phosphate groups, on each side. The opened DNA is used as a template and the rules of pairing between the different bases are followed (Figure 2). In this way two molecules of DNA, each identical to the original, are produced. When the cell divides, each daughter cell has its own identical copy of the DNA molecule.

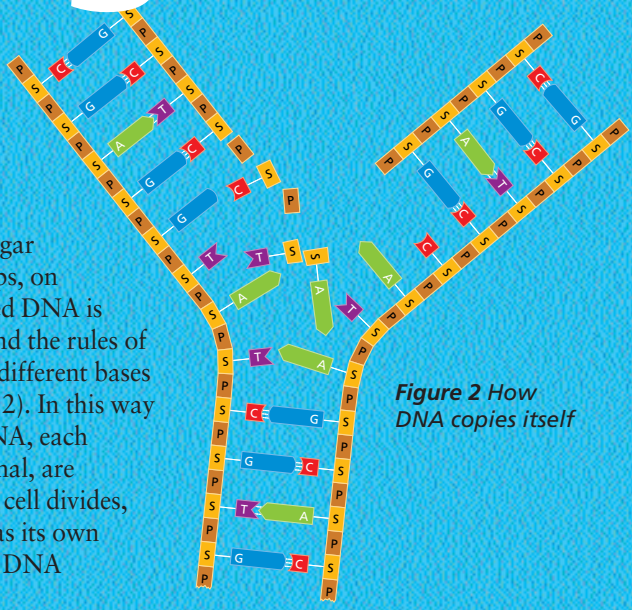


Figure 2 *How DNA copies itself*

In the last 50 years there has been a great leap in what we understand about DNA. Some of the other articles in this issue of CATALYST describe these advances.

CATALYST also contains plenty of other fascinating science to support you in your GCSE. Improve your Grade appears in every issue and aims to help you directly with your exams and coursework. In this issue it looks at variables in experiments. There are also articles covering all areas of GCSE science.

Nigel Collins

ROSALIND MIST

DNA fingerprinting

GCSE key words
DNA
Clone
Selective breeding

When Dolly the sheep was born in 1996, it was announced that she was a 'clone' from a female adult sheep. To prove this, the scientists who created Dolly had to have genetic profiling or fingerprinting done by another, independent, group of scientists. This article describes how it was done, and explains other uses of the technique.

Alfred Pasieka/SPL

Dolly with Ian Wilmut who led the team that created her

Topham

Have you ever wondered how the scientists who bred Dolly the sheep could be sure that she was a clone? They had to convince the scientific community that no mistakes had been made, and it was therefore important that more than one group of scientists tested Dolly.

Any organism produced by sexual reproduction is genetically unique (with the exception of identical twins) and this means the DNA packed into the chromosomes is different in each individual. Some years ago Sir Alec Jeffreys, a professor at the University of Leicester, developed a method for spotting unique features of an organism's DNA. His method formed the basis for genetic fingerprinting or profiling (see Box 1 and Figure 1), which was the technique used to check Dolly's genetic origin.

HOW DOLLY WAS TESTED

Human DNA is unique to the individual, but what about sheep DNA? The first thing to do was to compare DNA samples from several sheep to see whether sheep DNA is also unique. The team compared the DNA fingerprints of 12 different sheep. About 38% of the DNA bands in any two animals were similar. If the match between samples from Dolly and the donor ewe was much better than this, they were very likely to be related.

Dr Esther Signer, a member of the group at the University of Leicester which tested Dolly, said, 'We compared the DNA found in Dolly's blood with the DNA from the mammary cells of the donor ewe and with a sample of the original cells that had been implanted in the surrogate mother.' They found that there were no differences between the three samples. 'The chances of two unrelated sheep having such a good match were estimated at about 1 in 1600 million,' said Esther.

The group also had to consider the possibility that Dolly was a daughter sheep rather than a clone. Dolly's mother was pregnant when the mammary cells were taken. There was a small possibility that a foetal cell had travelled through the blood system to her mammary glands and been collected there by the scientists.

If she was a daughter, Dolly would have had about half her DNA from the donor sheep and the other half from a father sheep. As Dolly's DNA fingerprint was so similar to that of the donor ewe, Esther's team had to work out what the chance was that a 'normal' offspring could have had such a great resemblance. 'The probability was very small, 1 in 3 million,' said Esther.

NOT JUST SHEEP AND PEOPLE

DNA fingerprinting isn't just used to test the paternity of people, or even of 'cloned' sheep. The technique has been used for other purposes too.

Suppose you want to breed a prize pig, or one that will taste better than other pigs. You have a

BOX 1 GENETIC PROFILING

Everyone's DNA has the same molecular structures. The differences between the DNA of individual organisms lie in the order of the base pairs. There are so many millions of base pairs in DNA that every person has a different sequence except identical twins, nature's 'clones'.

Since these patterns are unique to an individual they are called DNA fingerprints. Unlike traditional fingerprints they can also show whether people are related.

Each person, animal or plant can be identified solely by the sequence of their base pairs. However, because there are so many millions of base pairs, this task would be very time-consuming. Alec Jeffreys found a short-cut making use of repeating patterns in DNA.

These patterns do not give an individual 'fingerprint', but they are able to show whether two DNA samples are from the same person, related people, or non-related people (or other organisms). Scientists use a small number of sequences of DNA that are known to vary a great deal among individuals, and analyse these to get a certain probability of a match.

Figure 1 *How DNA fingerprinting works*

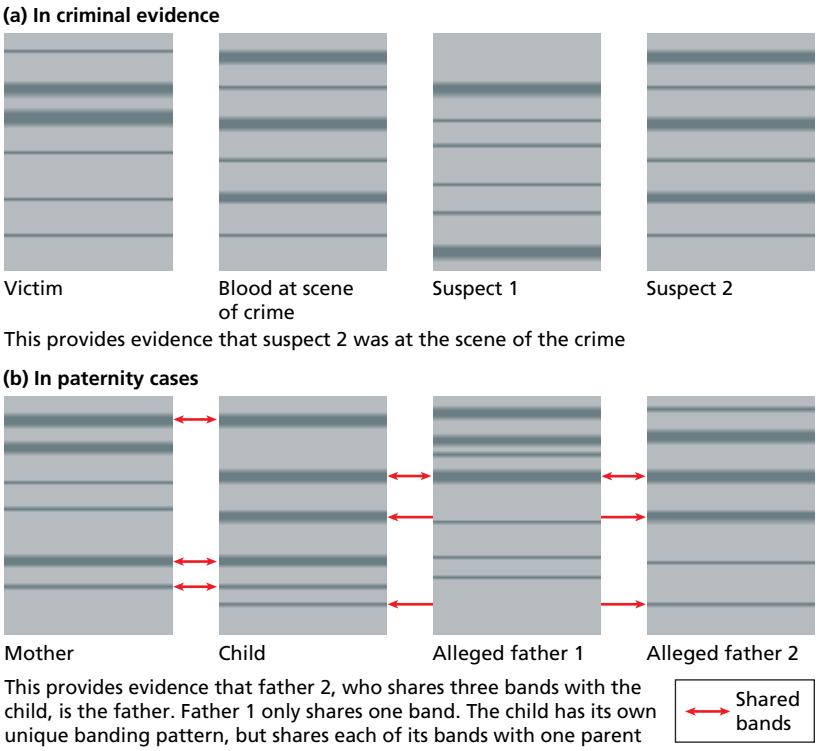
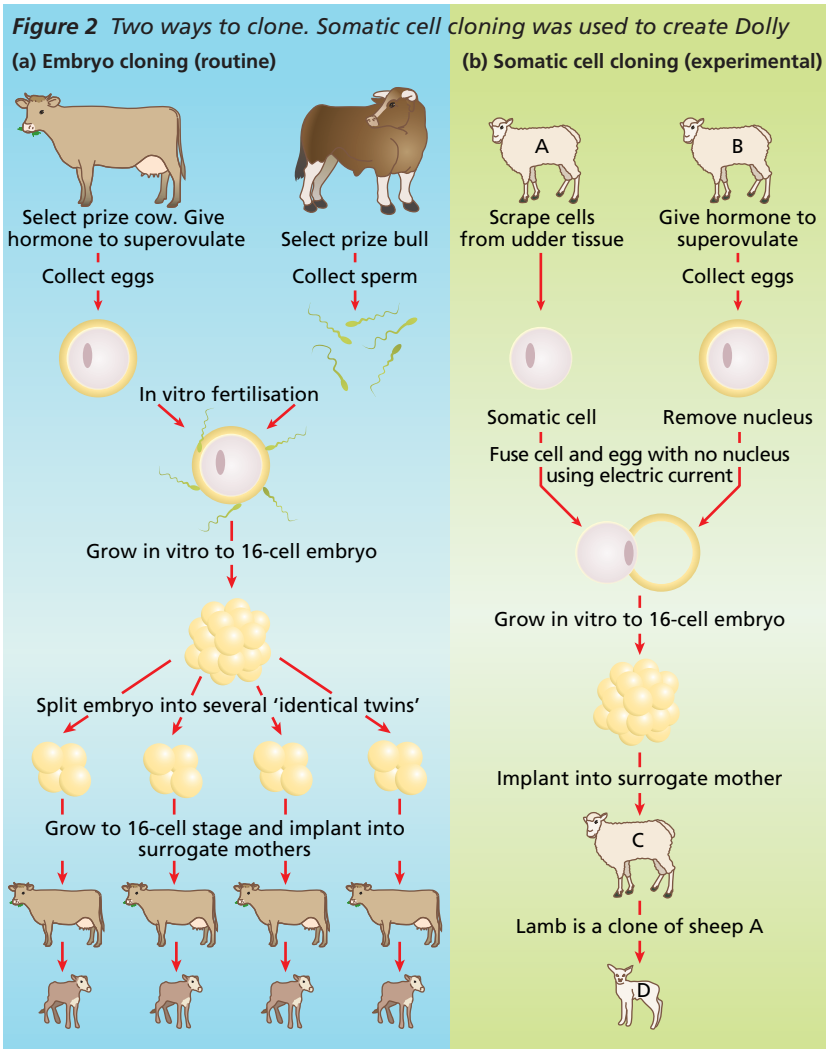
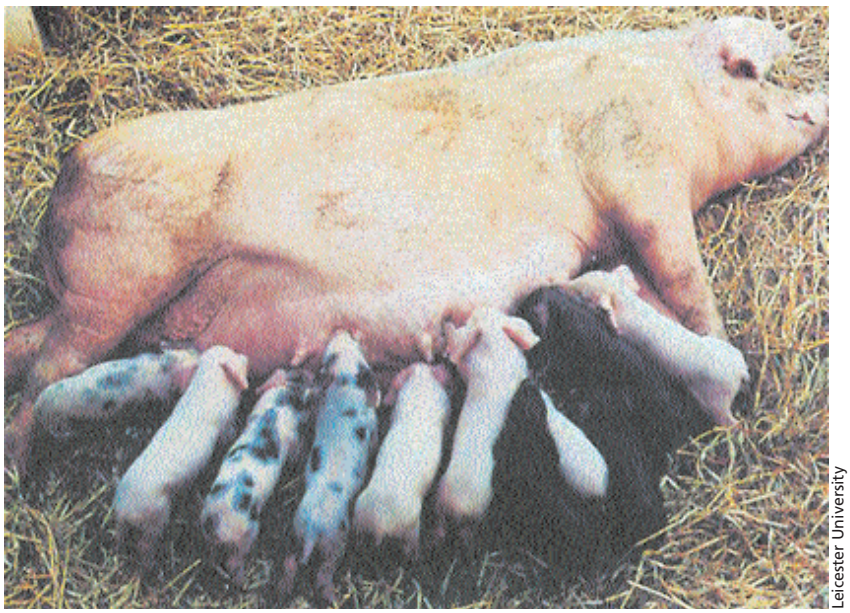


Figure 2 *Two ways to clone. Somatic cell cloning was used to create Dolly*



Leicester University



Leicester University

***Left**: Esther Signer's work on DNA has involved her in identifying the parentage of piglets (**below**) as well as that of Dolly, and of peregrine falcons*

sow, and you need a prize-winning father. You take your sow to be mated with the prize boar (or maybe you opt for artificial insemination). Sixteen weeks later, a fine litter of piglets is born. They grow up. However, as they grow, you are not quite sure about them — they are not what you expected. You begin to wonder whether the father was the boar you paid to use. What can you do?

Well, you can call in a DNA fingerprinting expert and see what they say. In the case of the pigs, Dr Signer discovered that somehow there had been a mix-up at the pig-breeding company. The test tubes containing the semen had not been labelled correctly, and the wrong pig had fathered the litter. You might think this is a lot of trouble to go to for a pig, but getting a prime porker can be an expensive business!

FORENSIC SCIENCE

You probably know that the police can use DNA as part of their forensic evidence in crime scenes, but did you know that they have used it to trap wild-bird smugglers and breeders?

In 2000, the RSPB (Royal Society for the Protection of Birds) suspected that some peregrine falcons it had found at a man's house were not all they seemed. The peregrine falcon is a protected species, but the man claimed his birds had been bred in captivity, and were not illegal wild birds. The RSPB suspected that he had taken them from the wild and called in the team at Leicester to investigate.

Esther got to work with the fingerprinting techniques and compared the DNA of the questionable chicks to that of the adult birds the breeder claimed to be the parents. The chick DNA profiles should have been a mix of the bands seen in their parents' profiles, but — you've guessed it — the matches were far from perfect. Using this evidence, the man was convicted under CITES (Control on International Trade in Endangered Species).

This was one of many cases brought by the RSPB using DNA evidence which have had a dramatic effect. 'Before DNA fingerprinting, we couldn't prove that birds were not reared in captivity,' said the RSPB spokesman. 'Thanks to DNA typing, there has been a significant drop in the numbers of people taking birds from the wild'.

***Rosalind Mist** runs sciZmic, a national network of science clubs based at Science Centres such as Explore@Bristol. **Esther Signer** is a geneticist at the University of Leicester.*

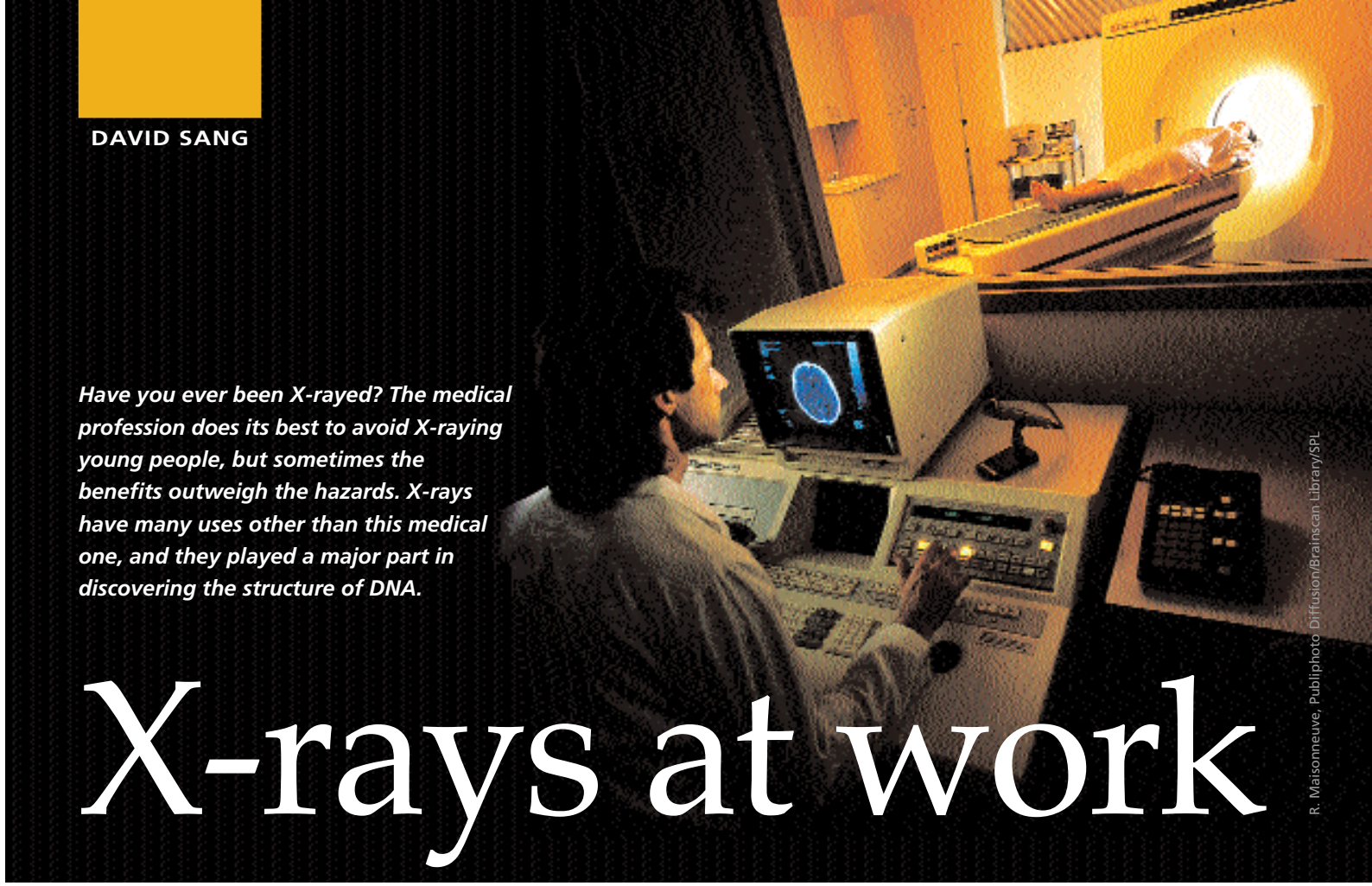
BOX 2 WEBSITES

http://www.dnai.org provides all sorts of animations, including those associated with the polymerase chain reaction used to copy DNA (see back cover), as well as video clips and historical background on many genetic topics. DNA fingerprinting itself is soon to be added to the site.

http://www.rspb.org.uk has information on how to watch and learn about wild birds.

http://www.cites.int explains CITES and the work it does to prevent trade in endangered species.

DAVID SANG



R. Maisonneuve, Publiphoto Diffusion/Brainscan Library/SPL

Have you ever been X-rayed? The medical profession does its best to avoid X-raying young people, but sometimes the benefits outweigh the hazards. X-rays have many uses other than this medical one, and they played a major part in discovering the structure of DNA.

X-rays at work

X-rays were discovered over 100 years ago by Wilhelm Röntgen. German-born, he was a professor of physics at the University of Würzburg, and that's where he made his big discovery. He was carrying out experiments using a cathode ray tube — an evacuated glass tube, rather like a television tube. It contains two metal electrodes which are connected to the positive and negative terminals of a high-voltage supply.

Switch on the supply, and a small current flows between the electrodes. Electrons escape from the cathode and are attracted to the anode. The space between the electrodes glows with light. This is because the vacuum in the tube is not perfect; it contains air at low pressure. Put a different gas in the tube and it glows a different colour.

STRANGE RAYS

On 8 November 1895, Röntgen was working with a cathode ray tube covered in light-tight black paper when he noticed something strange. Nearby was a screen coated with fluorescent paint, the sort of screen that is used for showing up invisible ultraviolet radiation. When Röntgen switched on his vacuum tube, the screen glowed. Some rays — some type of radiation — must be passing through the black paper and reaching the screen.

Röntgen soon established that the rays from his tube could pass through a wooden plank and even through metal sheets. And when he put his hand in the path of the rays, he noted that, 'flesh is very transparent, while bones are fairly opaque.' Röntgen could scarcely believe his results, but after weeks of concentrated work, he was able to publish three

BOX 1 MAKING X-RAYS

In any X-ray tube (Figure 1) there are two electrodes. A beam of electrons is produced by the **cathode**; the electrons crash into the metal **anode**, and their kinetic energy is transformed into X-rays.

The bigger the voltage between the two electrodes, the more the electrons are accelerated and the faster they are moving when they hit the cathode. High voltages produce high-energy, short-wavelength X-rays.

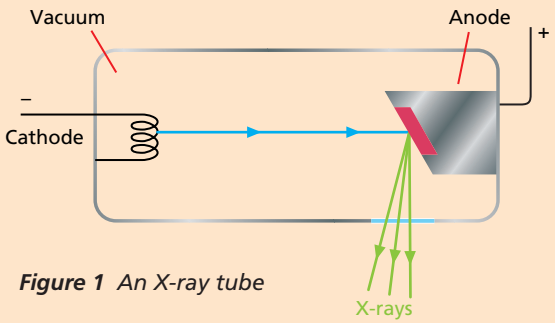
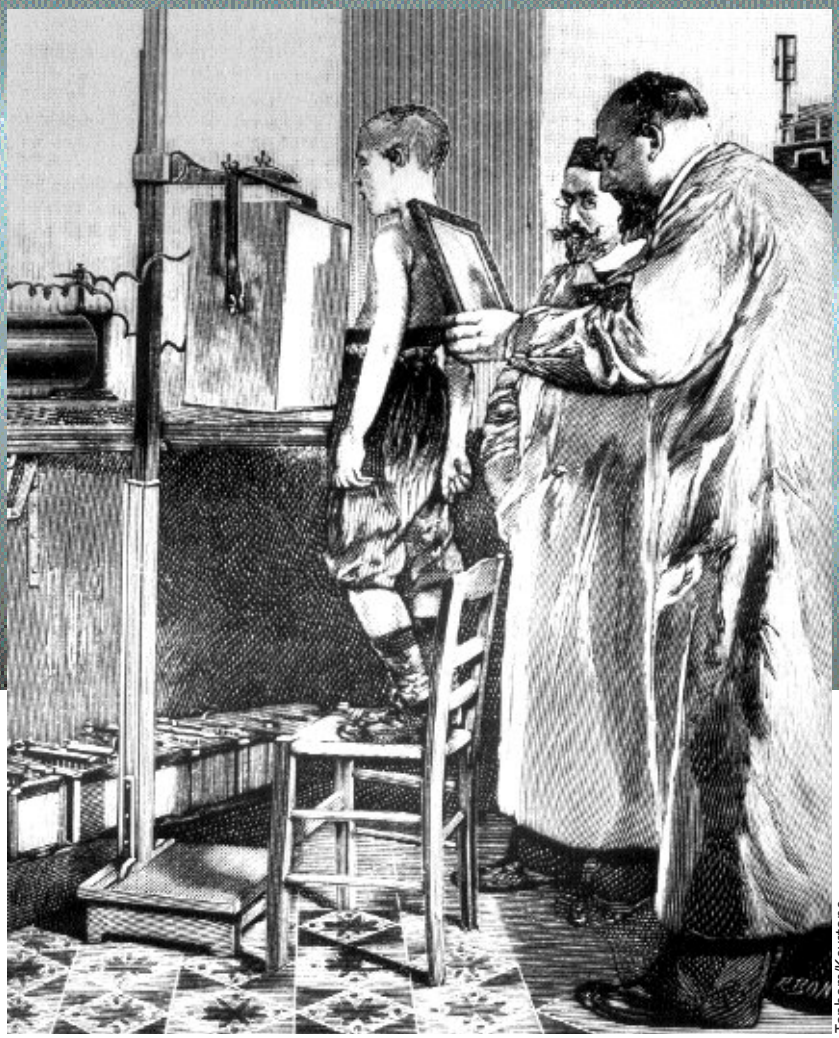


Figure 1 *An X-ray tube*

Above: *A CT scanner is a sophisticated X-ray machine. The source travels round the patient, producing images from different angles which are combined to form a three-dimensional view. CT stands for computed tomography ('slice-drawing'). Magnetic resonance imaging (MRI) uses radio waves instead of X-rays*

GCSE key words
X-ray
DNA
Ionising radiation
Electromagnetic spectrum



Topham/Keystone

Above: *Few safety precautions were taken by early users of X-rays*

So that he could devote more time to his studies, Röntgen had his bed moved into the laboratory, and his wife Bertha brought him his meals there.

papers outlining his discovery. He called his invisible radiation X-rays, because X stands for the unknown.

GOING PUBLIC

Wilhelm Röntgen's discovery caused a sensation. X-ray tubes were manufactured in large quantities, and many were sold to 'hobby scientists' to use at home. Public lectures and demonstrations were widely performed, and members of the public could volunteer to have themselves X-rayed in front of an audience. Röntgen himself shied away from this type of activity and gave only one public lecture.

X-rays found an immediate use in medicine, for examining fractured bones before operations. The rays pass through some materials more readily than others. We say that flesh transmits X-rays more readily than bone; bone is an absorber of X-rays. The thicker the bone, the more the X-rays are absorbed.

SPL

BOX 2 EXTENDING THE SPECTRUM

By the middle of the nineteenth century, scientists knew that the spectrum of light could be extended at either end to include two other radiations invisible to our eyes: infrared, which can be detected by its heating effect, and ultraviolet, detected using photographic film or a fluorescent screen (Figure 2). Wilhelm Röntgen's discovery of X-rays extended the spectrum further still.

Light, infrared, ultraviolet and X-rays are all types of electromagnetic radiation, and they all travel at the same speed through empty space. Beyond the ultraviolet region of the spectrum lie X-rays and gamma rays. X-rays travel as waves with a shorter wavelength than ultraviolet, while gamma rays (from radioactive substances) have even shorter wavelengths.

Figure 2 *Extending the electromagnetic spectrum*

Wavelength	Radio waves/ microwaves	Infrared	Visible light	Ultraviolet	X-rays	Gamma-rays
Discoverer	Hertz	Herschel		E. Becquerel	Röntgen	H. Becquerel
Date	1887	1799		1839	1895	1896

BOX 3 INSIDE CRYSTALS

In the early 1950s, Rosalind Franklin used X-rays to try to discover the structure of DNA. We can think of X-rays as waves with wavelengths similar to the size of atoms. This means that a beam of X-rays will be diffracted (spread out) a lot when it passes between atoms.

In a crystal, atoms or molecules are regularly spaced. They make an excellent arrangement for diffracting X-rays. What happens is this: a narrow beam of X-rays is directed at a crystal. As the rays pass between pairs of atoms, they spread out — that's **diffraction** (Figure 3). In certain directions, neighbouring waves are in step and reinforce each other (constructive interference). In other directions, they are out of step and cancel each other out (destructive interference). Strong beams of X-rays emerge in particular directions, and are detected, appearing as spots on a photographic film.

This is the basis of **X-ray crystallography**. If the atoms are close together, the spots on the film are further apart, so the spacing of the atoms can be found (Figure 4). In addition, different arrangements of atoms give different patterns of spots.

Rosalind Franklin was lucky. DNA molecules are large and difficult to crystallise, and they tend to break up when exposed to energetic X-rays. She was able to make fibres of DNA which proved to be robust enough to give clear X-ray photographs. She showed that DNA had a spiral structure and she was able to work out the length of each turn of the helix.

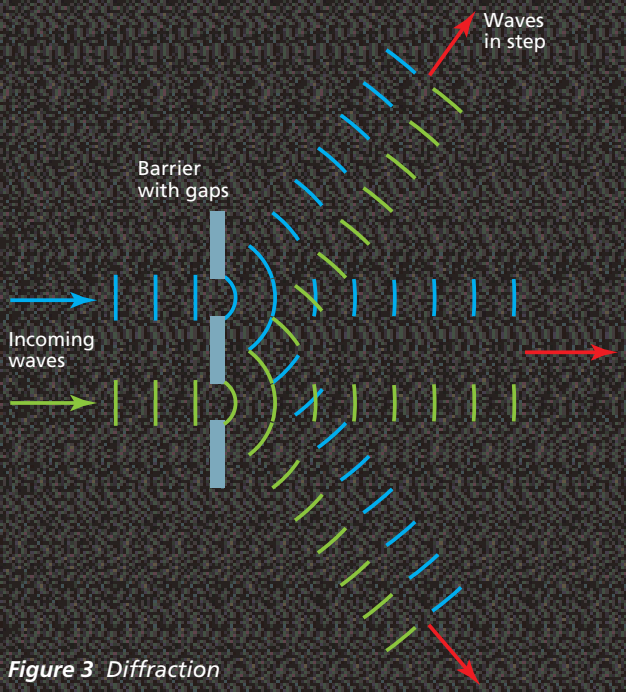


Figure 3 Diffraction

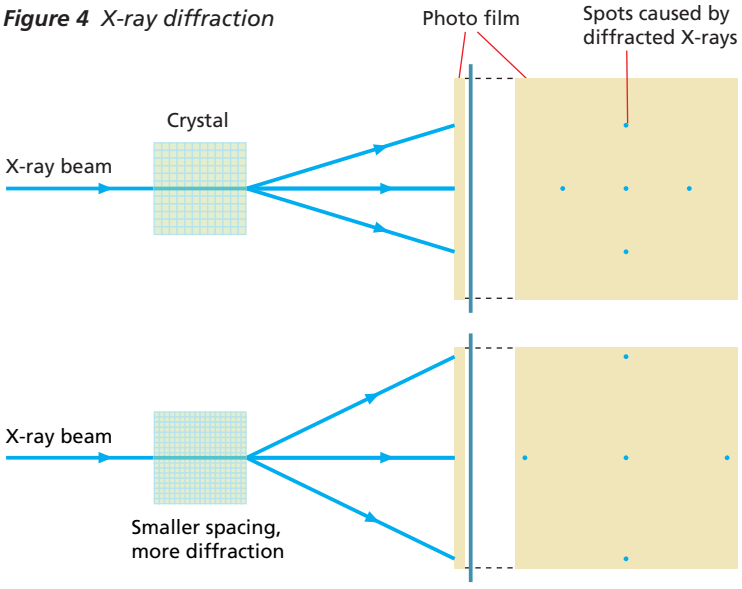


Figure 4 X-ray diffraction

X-RAY HAZARDS

The hazards of X-rays were not recognised soon enough. Medical staff regularly exposed their own hands to X-rays, to judge the length of time needed to get a good image. They often suffered reddening and even burns, but they never imagined the further harm they might be doing themselves.

X-rays (like gamma and ultraviolet rays) are a type of ionising radiation; that is, a ray may strike a molecule and cause it to become ionised. An electron may be knocked from the molecule, so that it becomes a positive ion; or the molecule may be split into two parts, one positive and the other negative. This can cause a **mutation**, a change in the inherited material, DNA.

If DNA is mutated, it may code for faulty proteins, including faulty enzymes. The mechanisms controlling a cell's normal development and division may be damaged and the result can be cancer. Early radiologists had an increased risk of leukaemia and skin cancer; many had to have fingers amputated.

Today, conditions for the use of X-rays are much more controlled. The dentist retreats to a safe distance when X-raying your teeth. In hospitals, technicians work behind lead-lined screens. The use of intensifying screens and electronic detectors also means that much lower doses of X-rays can be used.

Doctors try to avoid X-raying young people whose cells are developing and more vulnerable to damage. For elderly people, the situation is different. An X-ray may quickly reveal a problem without the need for invasive surgery. The risk to an older person is smaller, as they are less likely to live long enough for a cancer to develop. This is an interesting example of how benefits and costs can be balanced. It can be done only because we know about the possible consequences of exposing different tissues to different doses of X-rays.

Röntgen was lucky. He avoided the hazards of X-rays. He designed a metal darkroom in which he worked; his X-ray tube was outside, and the X-rays entered through a small hole. The metal walls protected him. He died aged 78 in 1923.

David Sang writes textbooks and is an editor of CATALYST.

Rosalind Franklin

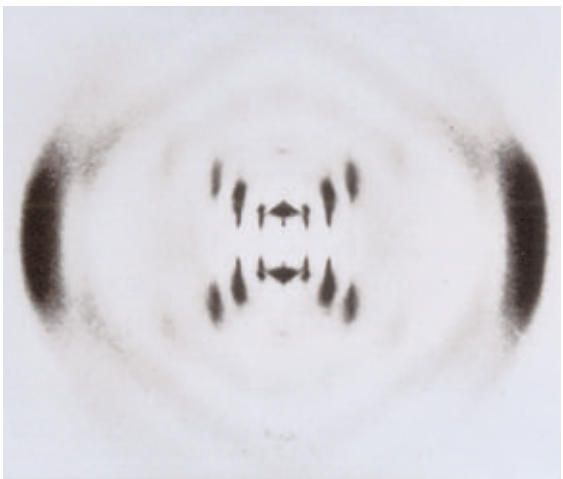
Rosalind in Paris, 1950

National Portrait Gallery, London

Rosalind Franklin was responsible for much of the research work that led to the discovery of the structure of deoxyribonucleic acid (DNA) in 1953. She died of ovarian cancer in 1958, at the age of 37, and did not receive the credit she deserved. This article tells her story.

Rosalind Franklin was born on 25 July 1920, the daughter of a London banker and the youngest of six children. She attended St Paul's Girls' School in West Kensington and soon found that she excelled in science. Her father wanted her to become a social worker but he relented, and in 1938 she entered Newnham College, Cambridge to study chemistry. Although she graduated in 1941, she stayed on for another year on a graduate fellowship, working on polymerisation reactions.

In 1942 Rosalind started work at the British Coal Utilisation Research Association where she made fundamental studies of the structures of carbon and graphite. She also became an air-raid warden, helping to safeguard London against German bombing attacks during the Second World War. Her work on the microstructures of carbon formed the basis for her doctorate in physical chemistry which she gained from Cambridge in 1945.



SPL

***Right:** Rosalind's X-ray diffraction photos of DNA were vital in the discovery of its structure*

X-RAY WORK

After her doctorate, Rosalind spent 3 years working in Paris where she learned X-ray diffraction. This technique involves looking at the image of a crystal under an X-ray beam (see Box 3, page 7). The location of atoms in the crystal can be worked out, and the precise structure mapped.

In 1951 Rosalind returned to London to work as a research associate in John Randall's laboratory in King's College. Here her path crossed with that of Maurice Wilkins. Both Rosalind and Maurice were working on finding out the structure of DNA, but Maurice always treated Rosalind as if she was a technical assistant, and not his equal, which she undoubtedly was.

Rosalind persisted in the DNA project and made crucial contributions to solving the structure of DNA. She recognised that two forms of DNA exist, and indeed discovered the second form. It was she

who placed the phosphate backbone on the *outside* of the molecule, contrary to what many of her contemporaries believed at the time. Her X-ray diffraction patterns were of excellent quality, and the scientist J. D. Bernal commented:

> As a scientist Miss Franklin was distinguished by extreme clarity and perfection in everything she undertook. Her photographs are amongst the most beautiful X-ray photographs of any substance ever taken. Their excellence was the fruit of extreme care in preparation and mounting of the specimens as well as in the taking of the photographs.

SOLVING THE STRUCTURE

Maurice Wilkins showed one of Rosalind's photographs of the B form of DNA to two researchers working in Cambridge in the early 1950s — Francis Crick and James Watson. This was done without her permission and it enabled Crick and Watson to finally formulate the structure of DNA. Crick and Watson published an article in the science journal *Nature* almost straight away (see page 1). Rosalind published her work as a supporting article in the same issue.

In 1953 Rosalind moved to J. D. Bernal's laboratory at Birkbeck College in London where she worked on the tobacco mosaic virus, and also on the polio virus. She went on a number of lecture tours of the USA.

AN UNTIMELY DEATH

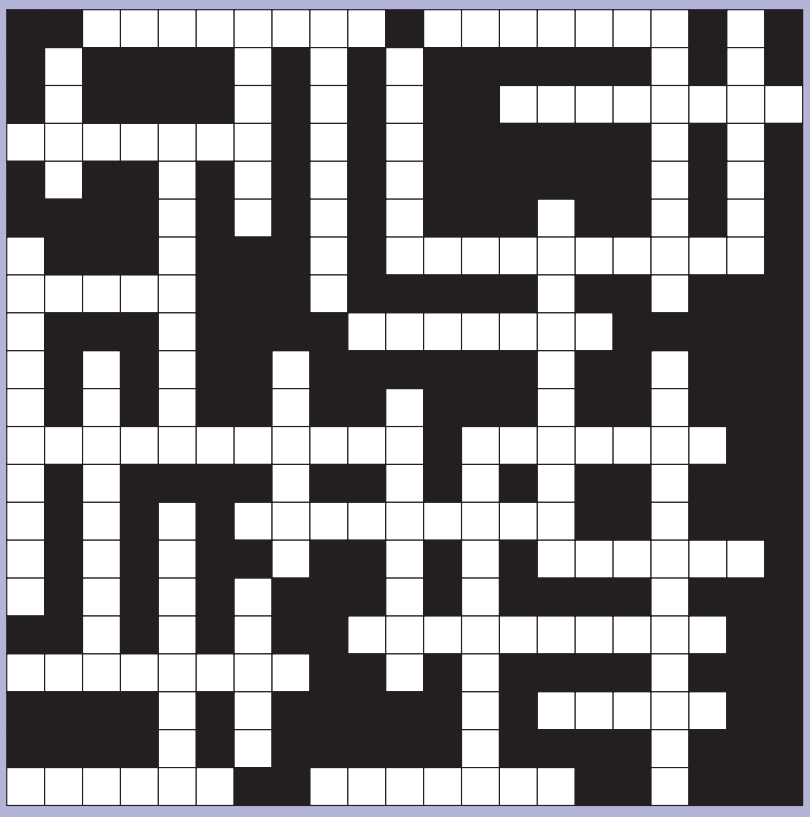
In 1956 Rosalind suffered abdominal pains which were diagnosed as ovarian cancer. She continued to work through her treatment and tried to continue her life and research as normal. Sadly, she died of cancer on 16 April 1958.

Maurice Wilkins, Francis Crick and James Watson received the Nobel prize for physiology and medicine in 1962 for discovering the structure of DNA. Nobel prizes are never awarded posthumously and never to more than three people at a time. What would have happened had Rosalind lived we shall never know. However it is a shame that Rosalind Franklin did not receive full credit for her essential role in solving the structure of DNA, either in her lifetime or, to a great extent, after her death.

David Moore teaches chemistry at St Edward's School in Oxford and is an editor of CATALYST.

An unbiased view of Rosalind Franklin's life and times can be found in the recent biography, *Rosalind Franklin: the Dark Lady of DNA*, by Brenda Maddox, published by HarperCollins, 2002.

Wordsquare



Use the words below, from Green Solutions on pages 11–13, to complete the wordsquare.

4-LETTER WORDS
SMOG

5-LETTER WORDS
FLUID
IONIC
TOXIC

6-LETTER WORDS
CARBON
EFFECT
FORCES
GLOBAL
STARCH

7-LETTER WORDS
DENSITY
HAZARDS
MONOMER
PATIENT
POLYMER
PRODUCT

8-LETTER WORDS
COVALENT
INDUSTRY
MENISCUS
PRESSURE
REACTION
RECYCLED
SOLUTION

9-LETTER WORDS
MEDICINES
MOLECULES
POLLUTION

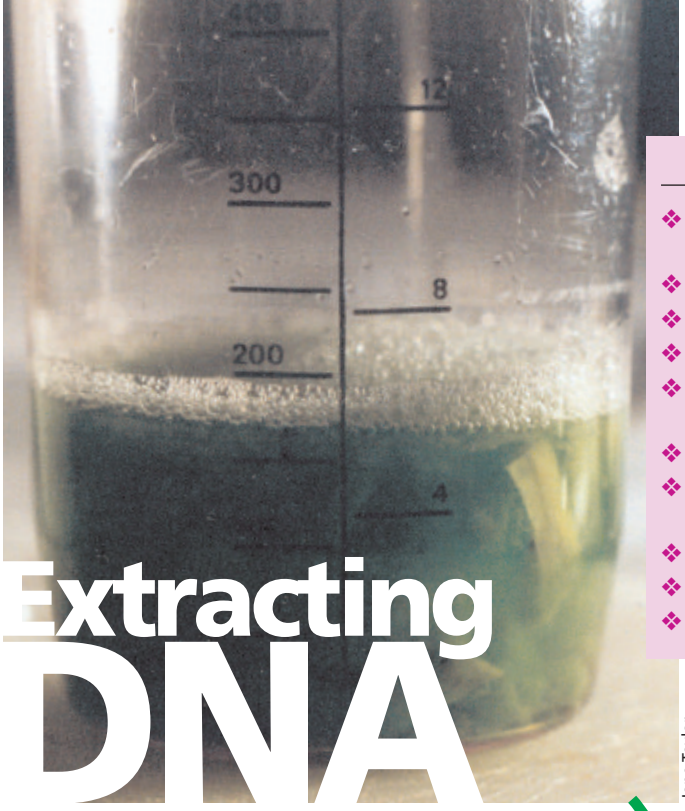
10-LETTER WORDS
ATMOSPHERE
GREENHOUSE
LABORATORY
NOTTINGHAM
POLYETHENE

11-LETTER WORDS
POLYSTYRENE

12-LETTER WORDS
INCINERATION

Answers on page 18.

TRY this

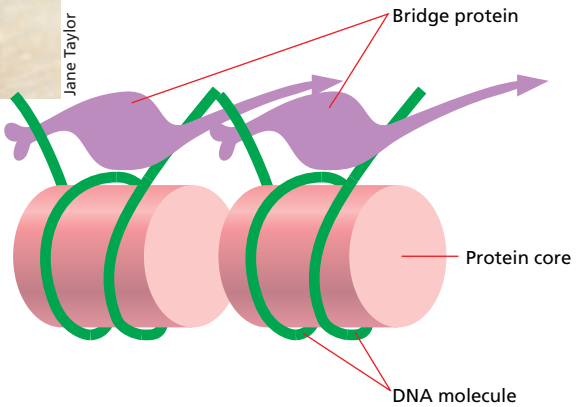


Extracting DNA

Right: *Extracting DNA from kiwi fruit*

WHAT YOU NEED

- ½ mug of wheatgerm or 1 kiwi fruit or 1 small onion
- water
- salt
- juice from ½ lemon
- pineapple juice from a tin of pineapple in natural juices
- washing-up liquid
- ice-cold meths or strong colourless spirits such as gin or white brandy
- fine sieve
- beaker
- measuring jug
- teaspoon
- mug
- liquidiser



Jane Taylor

Figure 1 *DNA is a giant molecule inside cells. It is packaged with proteins called histones, to take up little space yet allow the genes to work*

You can easily extract DNA from plants in a two-stage process, first removing DNA from cells, then precipitating it. We have used these methods to obtain DNA from wheatgerm, kiwi fruit and onions. The technique to use depends on your starting material.

Safety: Alcohol is flammable, so no naked flames. And ask your parents before you use any! Chill the alcohol by standing it in ice. Do not put it in the freezer compartment.

MAKING THE EXTRACT

Wheatgerm

Mix 150 cm^3 (¼ pint) of water, 2 heaped teaspoons of salt and the lemon juice in the measuring jug. Add the wheatgerm and stir gently for 10 minutes. Sieve the mixture and *keep the pulp*. Dissolve ⅓ teaspoon of salt, 6 teaspoons of alcohol, 2 squirts of washing-up liquid and 1½ tablespoons of pineapple juice in 75 cm^3 water. Add the pulp and stir for 20 minutes to allow the DNA to be dissolved. Add 3 good teaspoons of salt and stir for a further 10 minutes. Leave the mixture to stand so the solid starts to settle. Pour the *murky liquid layer* into a glass— this contains the DNA.

Jane Taylor

Jane Taylor

DNA extracted from a kiwi fruit. It appears as fine threads in the layer of methylated spirits

Kiwi fruit or onion

Chop a peeled kiwi fruit or onion into small pieces. Add to a solution of 1½ heaped teaspoons of salt and 5 squirts of washing-up liquid in 150 cm^3 warm water. Stand the mixture in a bowl of warm water for 15 minutes, stirring gently. Mix in pineapple juice as before and liquidise for a few seconds, then strain into a beaker. Don't let the foam get into the final stage.

PRECIPITATING THE DNA

Pour a small amount of DNA extract into a small glass, then carefully add three times as much ice-cold alcohol down the side of the glass to form a layer on top of the extract. DNA will appear as fine white threads suspended in the alcohol during the next couple of minutes. You can lift it out with a glass 'swizzle-stick' if you are careful.

LUCINDA DUDD AND KELLY MORLEY

Green solutions

Many reactions take place in solution. Water is a familiar solvent but there are many others. In this article we explore some of these, including a rather surprising one — carbon dioxide — which is a non-polluting 'green' solvent.

GCSE key words
Solvent
Particle theory
Solution
Carbon dioxide

The chemical industry relies on solvents to produce many everyday products we take for granted, such as medicines and plastics. The covalent organic starting materials used in these products often will not dissolve in ionic water (just as oil and water don't mix — think of salad dressings). Instead of water, industry uses solvents called **volatile organic compounds** (VOCs).

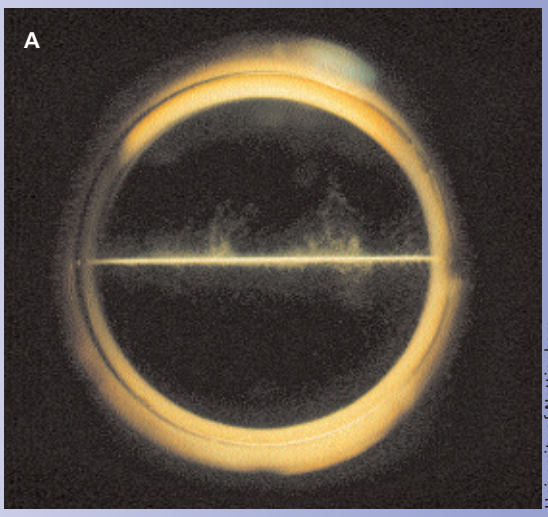
VOCs are derived from petrochemicals. This means they are produced from oil, which is a non-renewable resource. They are also associated with hazards, for example many are highly flammable. When the chemical reaction is complete, the solvent is either recycled or disposed of by incineration. But incineration can cause atmospheric pollution which can damage the environment and our health.

ALTERNATIVE SOLVENTS

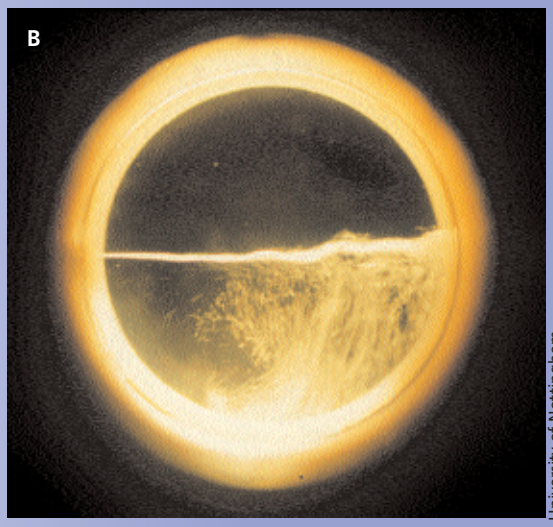
Researchers are trying to find alternatives to using VOCs as solvents. In our research at the University of Nottingham, we have used carbon dioxide as a solvent for many different types of chemistry and now industry is starting to use it too. Carbon dioxide is a gas at room temperature (it is a constituent of air and it makes the bubbles in carbonated drinks). It is not very good at dissolving organic compounds because gas molecules have weak attractive forces and cannot force the reactants together to produce a chemical reaction. However, if we compress carbon dioxide and heat it a little, it becomes a **supercritical fluid** and a much better solvent.

SUPERCRITICAL FLUIDS

Imagine boiling some water in a sealed container. As the water heats up, some of the molecules gain enough energy to break the attractive forces of the

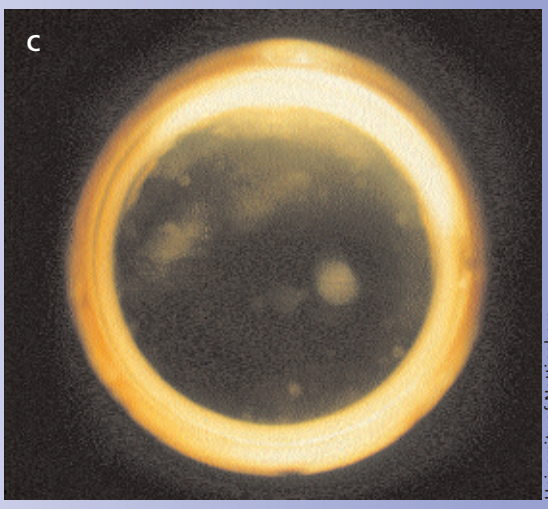


University of Nottingham



University of Nottingham

These three photographs show liquid being heated in a sealed vessel. You can see the liquid 'boiling' (B) and, at a certain temperature and pressure, it becomes a supercritical fluid (C)

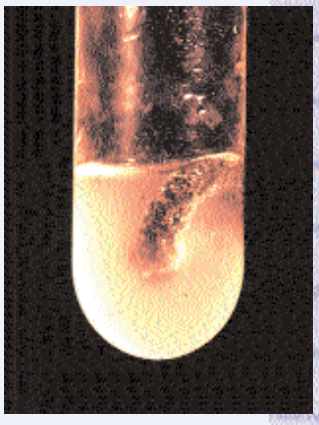


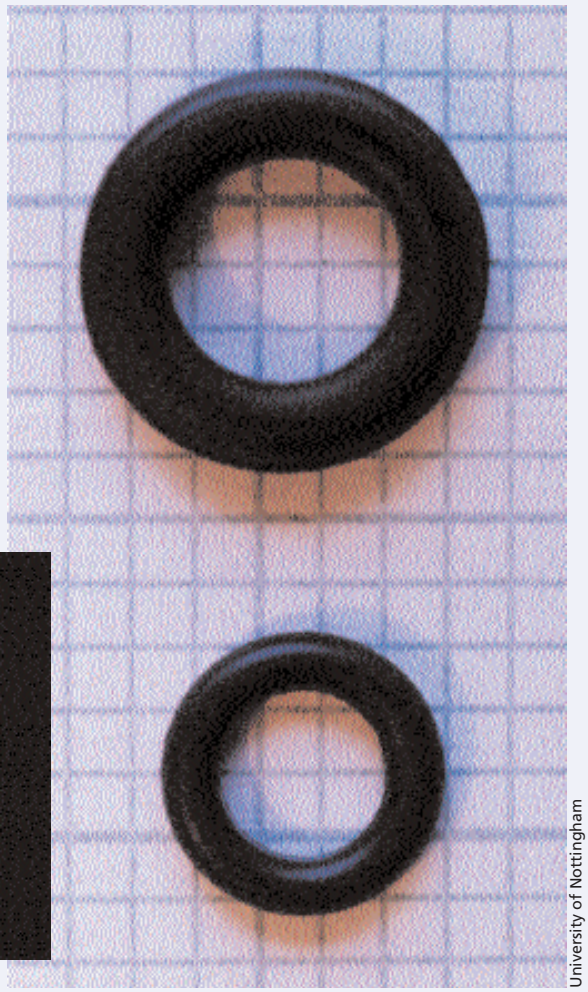
University of Nottingham

Never try heating anything in a sealed container yourself. A large amount of pressure will build up and burst open the container with lethal force. Researchers use specially designed stainless steel equipment and follow rigorous safety procedures.

Right: *Carbon dioxide has swelled the polymer ring at the top. Do you know how to test for the presence of carbon dioxide? The equation for the reaction is in Box 1*

Below: *The top ring has been put in limewater and as the carbon dioxide comes out of the ring (you can see the bubbles), it reacts to form calcium carbonate, which is a white solid and makes the water cloudy*





University of Nottingham

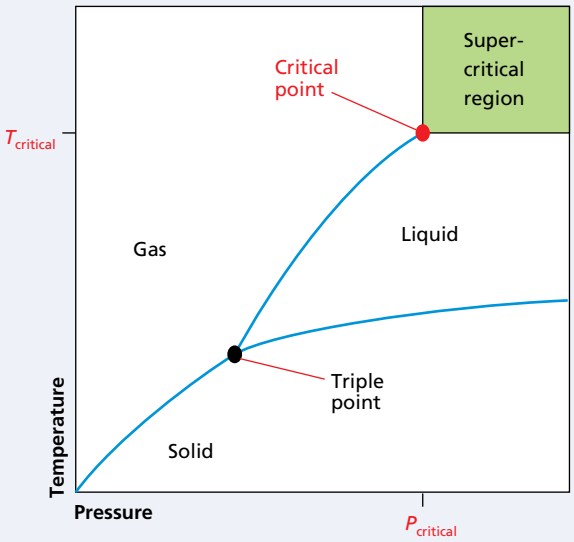


Figure 1 *A temperature–pressure plot for all matter. The triple point is the temperature and pressure at which a substance can exist in equilibrium in the solid, liquid and gaseous states. The triple point of water (0.01°C and 0.006 atmospheres) is used to calibrate thermometers. At the critical point, the liquid and gaseous states become identical. The critical temperature for carbon dioxide is 31°C and the pressure needs to be 73 times atmospheric pressure, while for water, the temperature needs to be 374°C and 218 atmospheres*

BOX 1 SOLVENTS

Chemical reactions are normally carried out in a solvent. Solvents are needed to mix the chemicals together and to remove any heat produced during the reaction. Without solvents, the reaction may give the wrong product or even blow up!

You will have used water as a solvent in the laboratory to dissolve ionic compounds. For example, calcium hydroxide is a solid and must be dissolved in water to help it react with carbon dioxide. This produces calcium carbonate, which is used to treat heartburn and indigestion. The equation for the reaction is

$Ca(OH)_2(aq) + CO_2 \rightarrow CaCO_3 + H_2O$

neighbouring molecules and so become a gas — the water starts to evaporate. The density of the liquid decreases (as molecules leave) and the density of the gas increases (as molecules enter) until liquid and gas have the same density. At this point, the meniscus (the surface separating the liquid and the gas) disappears and a different state of matter, called a supercritical fluid, is formed.

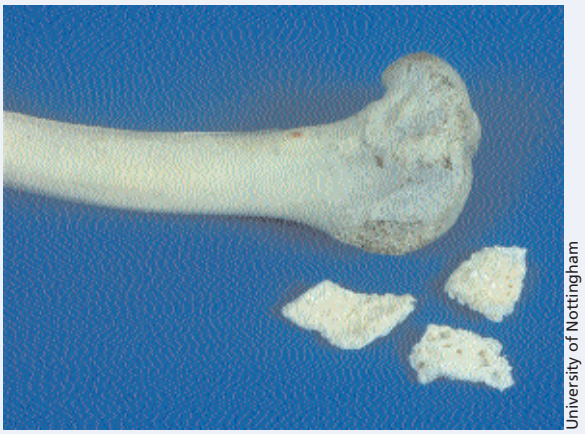
A supercritical fluid is a state of matter between a gas and a liquid and has some of the properties of both. For example, like a liquid, a supercritical fluid can dissolve things, but, like a gas, it can also diffuse through a solid. We can take advantage of these properties to make useful organic compounds that would be hard to make in conventional solvents.

POLYMER PROCESSING

A **polymer** is a chain of repeating units, called **monomers**. Polymers are all around you, from your baked potato (starch) to everyday plastics such as those in carrier bags (polyethene) or protective packaging (polystyrene). Using supercritical carbon dioxide ($scCO_2$) to process polymers not only replaces harmful conventional solvents but also allows new types of polymers to be made. When a polymer and $scCO_2$ are mixed together, the carbon dioxide is forced into the polymer and causes the polymer to swell up like a balloon. Substances can then be added into the polymer.

One possible application of $scCO_2$ is in modifying the plastic used for artificial hip joints. If the material can be made harder, then it may last longer in the body. We are trying to achieve this by adding metal particles to the polymer using $scCO_2$. Another area of our research using $scCO_2$ involves mixing drugs into biodegradable polymers. These new products can be taken by the patient as a pill. The pill slowly degrades in the body and the drug is released in a controlled way over a set period of time.

Supercritical carbon dioxide is an ideal solvent for processing polymers for medical applications because, unlike liquid VOCs, the carbon dioxide gas



University of Nottingham

diffuses out of the polymer and leaves no toxic residues behind.

GREEN CHEMISTRY

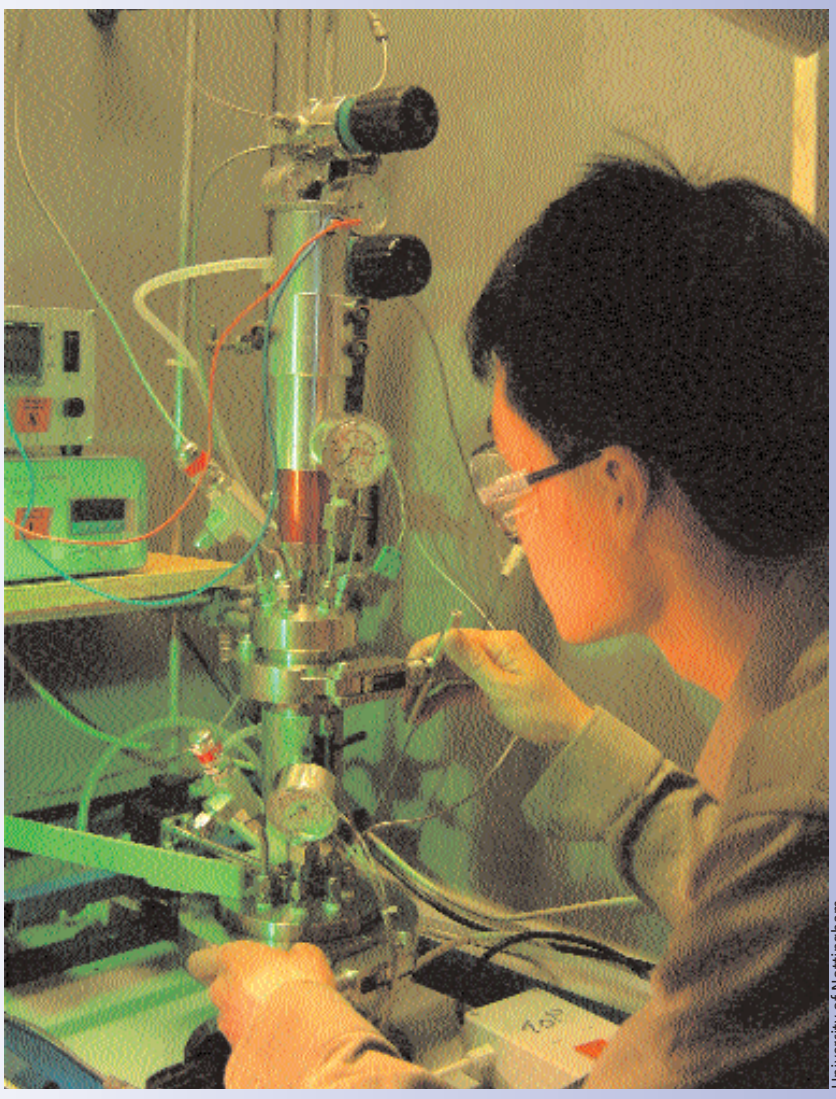
You might think carbon dioxide is not really a 'green' solvent because it is a so-called greenhouse gas, and increased levels of carbon dioxide in the atmosphere contribute to global warming. However, for our work we take carbon dioxide from companies that produce a lot of it (e.g. the brewing industry) and reuse it over and over again. We rarely emit it into the atmosphere. At the end of a reaction, the carbon dioxide is separated from the product by allowing it to expand to a gas and then compressing it again, ready for reuse.

Research carried out at the University of Nottingham is now being applied in industry to make new processes 'green' and existing processes 'greener'. Using green solvents is just one area of green chemistry, which aims to use less harmful chemicals and increase the efficiency of chemical processes.

For example, if the starting materials are harmful, green chemists try to find alternative starting materials to make the same product. If the product is harmful, green chemists try to find a different product that performs the same function. Using a catalyst speeds up the reaction so less energy is required — for this purpose organisms are being used to convert reactants to products, utilising their enzyme systems.

Industry is starting to realise that it is cheaper to use green chemistry than to comply with the ever-tightening regulations on the use and disposal of hazardous chemicals, and this is good news for the environment. The University of Nottingham is the first in the UK to offer an undergraduate degree in green chemistry, so a new generation of students will be helping our planet become a healthier place to live!

Lucinda Dudd *is studying for a PhD in supercritical water at the University of Nottingham.* ***Kelly Morley*** *has completed her PhD and now works for the Green Chemistry Network promoting all areas of green chemistry.*



University of Nottingham

A researcher uses high-pressure equipment to process biodegradable polymers using supercritical carbon dioxide. When the pressure is released slowly, the carbon dioxide leaves holes or pores inside the polymer. Polymers of this type may, in the future, be used to support the growth of new cells, such as liver or bone cells, in damaged tissue in the body. The polymer degrades over time in the body, leaving behind the new tissue. The photograph on the left shows small pieces of our porous polymer that have supported the growth of bone cells in the laboratory

BOX 2 WEBSITES

Find out more about our supercritical research at:
http://www.nottingham.ac.uk/supercritical
and about biodegradable polymers, which support new tissue growth at:
http://www.nottingham.ac.uk/pharmacy/tissue-eng
Go to a cyber wonderland of polymer fun at:
http://www.psrc.usm.edu/macrog
More on green chemistry can be found at:
http://www.epa.gov/opptintr/greenchemistry and
http://www.chemsoc.org/networks/gcn
while for wider environmental issues look at:
http://www.environment-agency.gov.uk

Clockwise from top left:

The Orange Imaginarium at night

In Your amazing brain

Web of life

Rainforest path in the Botanical House

@Bristol offers all sorts of opportunities to explore science, technology and natural history, many of which are relevant to your GCSE science courses. It is made up of three main sections: Explore @ Bristol, Wildwalk @ Bristol and an IMAX theatre.

EXPLORE @ BRISTOL

Explore @ Bristol is one of a number of hands-on science centres which have been developed around Britain. We have described some of these in previous issues of CATALYST. Explore @ Bristol offers more than 170 interactive activities and experiences, many of which are hands-on and use a variety of multi-media techniques. There are four themes in Explore.

Your amazing brain explores the workings of the human brain. In *Brain matters* you can find out how you see, hear, think, learn and dream. In the *Brain and body* section you can measure your pulse, take your blood oxygen level and see if, by squeezing a pump, you can keep up with your heart's action. Other sections here include *Super senses* and the amazing *Sperm journey*.

In ***Move it*** there is a series of displays and hands-on activities which allow you to explore a host of technological inventions and the science underpinning them. *Move it* is divided into five sections, *Machines*, *Strong shapes*, *Water works*, *Power it* and *Dreams of flight*. There are activities where you can move water with pumps, pistons, levers and locks. You can build bridges, use pedal power to generate electricity, use pulley mechanisms to lift yourself in the air and see how easy and how difficult it can be to make things fly.

Natural forces is one of three sections in the ***Curiosity zone***, and here you can make lightning follow your hand, explore the worlds of hurricanes and whirlpools, learn about magnets, and move

BOX 1 WEBSITES

To find out more and to check on the daily programme of events log on to:
http://www.at-bristol.org.uk

Buying tickets for all three attractions works out cheaper than single tickets.

For some interesting illusions, some of which are on display in *Your amazing brain* go to:
http://www.at-bristol.org.uk/Optical/default.htm

David Barbour

Paul Box

There is also an IMAX cinema at @ Bristol.

The Orange Imaginarium is a 100-seat stainless steel planetarium with a variety of programmes.

magnetic fluids. You can even step inside a tornado.

In *Focus on light* you can bend, bounce, split and mix light, make shadows and investigate light's surprising properties. The *Sound space* lets you see some sound waves, feel vibrations shake your whole body, test your rhythm skills and change the sound of your own voice.

The fourth theme is ***Get connected***, which links you via digital communication technologies to various places around the planet. You can visit the barren landscape of the north pole or see what the weather is like down under in Australia. You can also explore image manipulation and try your hand at television programme making.

WILDWALK @ BRISTOL

Wildwalk @ Bristol takes you on a cleverly thought-out trip through the living world. Live plants and animals, with integrated multimedia images and sounds, are grouped around five themes.

Origins of life presents an overview of biodiversity in *Variety of life*, while in *Simple beginnings* you can observe the microscopic world of bacteria and the simple forms of life that teem in a drop of water. You can discover how these microscopic organisms changed the future of life on Earth, and why most of them do us much more good than harm.

Building bodies offers clues as to how these single cells evolved to become complex multicellular organisms. You can touch the fossil casts of animals which lived more than 500 million years ago and, in *Fishes*, meet their relatives that live in the seas today.

The next section, ***Plants on land***, is housed in the Botanical House. Here you can trace the development of plants from simple mosses and liverworts, through horsetails, ferns and conifers, to the flowering plants. A large part of the Botanical House is occupied by tropical rainforest plants, ranging from prehistoric cycads to exotic palms, flowers and grasses. You can experience a slice of the steamy tropics, with sound effects and various animals.

Wildwalk is home to over 150 species of animals from all the major animal groups on the planet. Many of them are exhibited in a wide range of displays in ***Animals on land***. *Insects* and *Arachnids* both have their own sections. *Up on all fours* is devoted to the vertebrates.

Living planet allows you to look more closely at ecosystems and communities from a variety of perspectives and locations around the globe, including the deep ocean, leaf litter on a forest floor and a seabird colony.

The relationship between people and the environment is finely balanced. ***People and the planet*** provides an insight into how we threaten our environment and ways in which we can ensure its sustainable future.

HOW TO GET THERE

@ Bristol is only 2 minutes' walk from the city centre.

JANE TAYLOR

Explaining inheritance

This year is a double anniversary in the hunt for an explanation of inheritance. The role of chromosomes was announced in 1903 and the structure of DNA was revealed in 1953.

Dr Yorgos Nikas/SPL

Above: *Human sperm on the surface of an ovum during fertilisation*

GCSE key idea
Scientists, like other people, are reluctant to give up explanations that served them well in the past.

Since earliest times people have speculated about how features pass from generation to generation. A theory of **pangenesis** prevailed for centuries, from the time of Hippocrates (about 400 BC) onwards. This theory supposed that all parts of the body contribute some form of essence to the generative fluid or 'seed' of the male or female parent and that the essences of the parents are transmitted to the offspring at conception.

SPERMIST OR OVIST?

Things did not change much until the sixteenth century. Then **Anthony von Leeuwenhoek** ground some simple lenses with which he found tiny creatures swimming in seminal fluid. Until then no one knew that humans produced **eggs** and **sperm cells**. Leeuwenhoek thought he saw miniature individuals coiled up inside sperm cells, and this contributed to the idea that babies were pre-formed in sperm.

On the other hand **Marcello Malpighi** in Italy had seen a tiny embryo in a hen's egg and thought the egg carried the pre-formed individual. Two camps formed. The **ovists** favoured the egg as the source of the offspring, while **spermists** favoured sperm.

Sea urchins were useful for research because their large eggs and sperm can be observed in a cool Petri dish in a laboratory.

THE BIOLOGY EXPLOSION

By the mid nineteenth century new achromatic lenses gave a much better magnification and resolution, so that **Theodor Schwann** could see cell structure in detail. He suggested that **cells** were the basic unit of life. **Oskar Hertwig** watched the external fertilisation of sea urchin eggs and saw that the sperm penetrated the egg. He believed that sperm and egg nuclei fuse during fertilisation. Inherited material must therefore lie somewhere within both egg and sperm cells.

In 1882 **Walther Flemming** used Perkin's mauve to stain the nucleus and noticed a coloured material inside it that he called **chromatin**. Chromatin changed into threadlike strings, which became known as chromosomes, when the cell divided. He observed chromosome movement during cell division, which he called **mitosis**, but he did not connect it to inheritance.

FINDING OUT ABOUT CHROMOSOMES

Theodor Boveri also investigated what happened during fertilisation and the early development of sea urchins. By 1887 he had seen that maturing eggs lose half their nuclear material and that the sperm and egg nuclei do not fuse during fertilisation. Instead each contributes half the **chromosomes** which together make the normal number found in the cell. He saw that chromosomes stay as whole structures through cell division and this gives the continuity needed to pass information on to new cells. The idea that chromosomes were important in inheritance was unpopular.

Ten years later **Hugo de Vries** rediscovered Mendel's work on inheritance (see CATALYST Vol. 12, No. 4). Boveri spotted the match between Mendel's laws and what was known about chromosomes and suggested that chromosomes transmit hereditary characteristics. **Walter Sutton**, working on cell development, came to the same conclusion at the same time. Another clue was found at around the same time when it was realised that cells in females always have two X chromosomes while male cells have an X and a Y. This linked a feature (gender) to differences in chromosomes.

The Boveri–Sutton chromosome theory was hotly debated until **Alfred Sturtevant**, a student in Morgan's genetics laboratory, settled it in 1915. He realised that five characteristics of the fruit fly *Drosophila melanogaster*, on which he had been working, could be inherited as a cluster. Three other clusters of features were identified, making four groups that correlated with *Drosophila*'s four pairs of chromosomes. Further analysis confirmed that hereditary factors were carried on the four pairs of chromosomes. Morgan suggested that genes were arranged in a line in the chromosomes.

BIOCHEMISTRY

The biochemists had been busy too. **Friedrich Miescher** investigated the composition of white blood-cell nuclei in 1869. He extracted a new substance from them that he called **nuclein**. Nuclein had a different composition from other molecules, with high phosphorus and nitrogen content, and it resisted protease digestion so it wasn't a protein. Miescher thought that nuclein might be a store for phosphorus. Two forms of nuclein were eventually found but their structure and function were a mystery. **Albrecht Kossel** showed that a form of nuclein found in thymus cells contained four nitrogen compounds: adenine, guanine, cytosine and thymine. Other researchers found some carbohydrate and phosphorus.

Phoebus Levene showed that nuclein was different from proteins. In 1909 he found that the carbohydrate in a form of nuclein from yeast is a sugar, ribose. It took until 1929 to identify the carbohydrate in the thymus form of nuclein as a sugar called **deoxyribose**. Levene suggested a simple structure for the newly named **ribonucleic** and **deoxyribonucleic acids** (RNA and DNA). He proposed that there were units containing one molecule of each of the four bases, joined together by sugar and phosphate, and that these units were linked to make a polymer with bases in the same order repeating down the length.

This idea of a repetitive structure was a stumbling block in the search for the basis of inheritance. Scientists looking for something that could convey vast quantities of complex inherited information thought the simple structure proposed for nucleic acids was inadequate. The more complex and infinite structural variety of proteins had much more potential to encode such a diversity of information. The smart money looked for genes among proteins.

WAS IT PROTEIN OR WAS IT NUCLEIC ACIDS?

The idea of proteins carrying genes was overturned in 1944. **Oswald T. Avery** and his colleagues had been puzzling for years over a substance from killed bacteria that could give new abilities to living bacteria. By 1936 they felt that it must be a nucleic acid. It was pure DNA, and a series of tests eliminated any alternatives. In 1952, **Alfred Hershey** and **Martha Chase** provided the final proof that DNA carries genes when they showed that the DNA in bacteriophages (viruses that attack bacteria) is the part that directs the making of new virus particles, including their protein coats (see Box 1 on page 18).

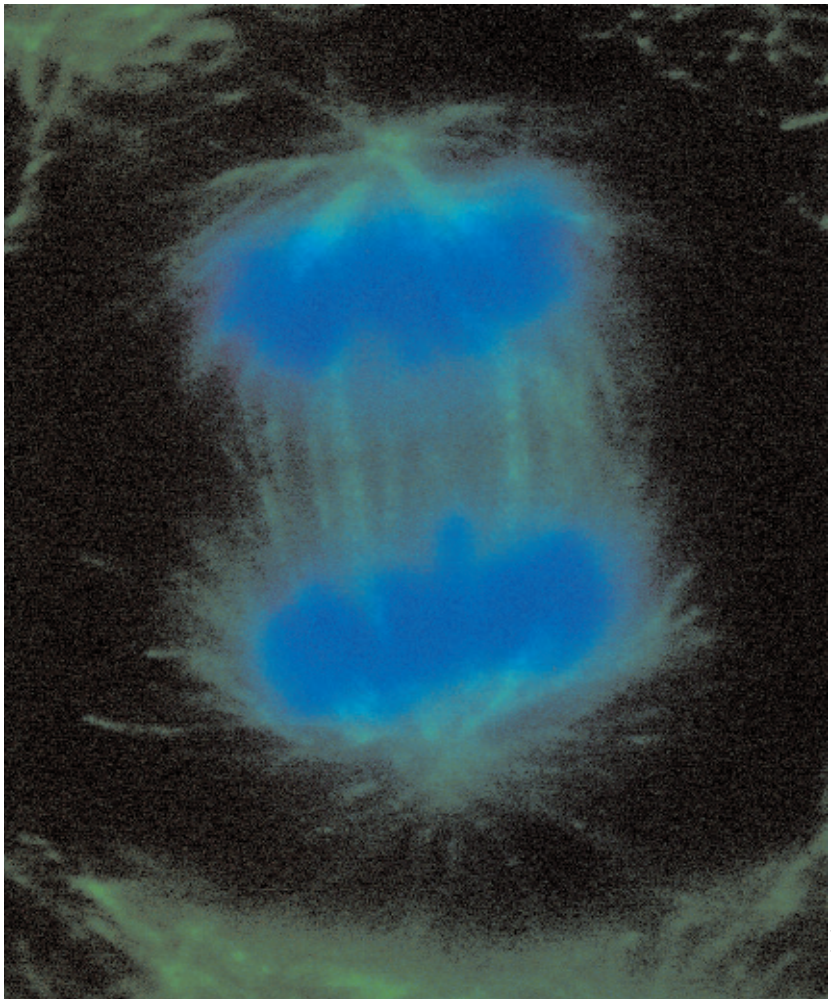
This work revived interest in DNA and people looked at its structure again. **Erwin Chargaff**'s observations disproved the idea of a simple DNA

***Drosophila melanogaster* is the tiny fruit fly often seen in summer.**

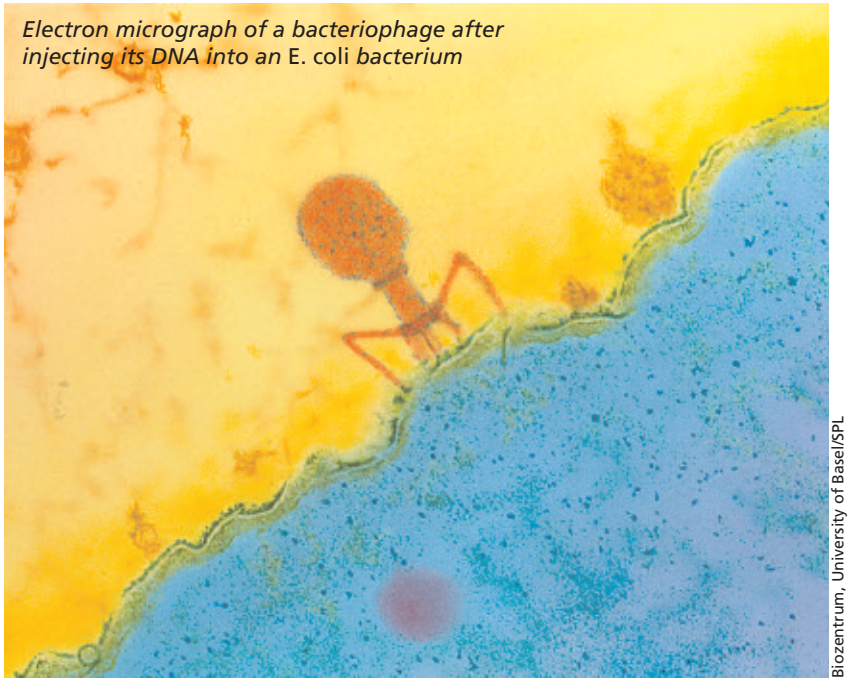
Gregor Mendel worked in relative isolation and his work was unknown to scientists working on inheritance for many years.

Many bacteria pass DNA into other bacteria. This is a normal way for bacteria which reproduce asexually to exchange genes.

***Below:** A cell during mitosis. The chromosomes (blue) have divided and are moving towards positions in which new nuclei will form*



J. W. Shuler/SPL



Electron micrograph of a bacteriophage after injecting its DNA into an E. coli *bacterium*

Biozentrum, University of Basel/SPL

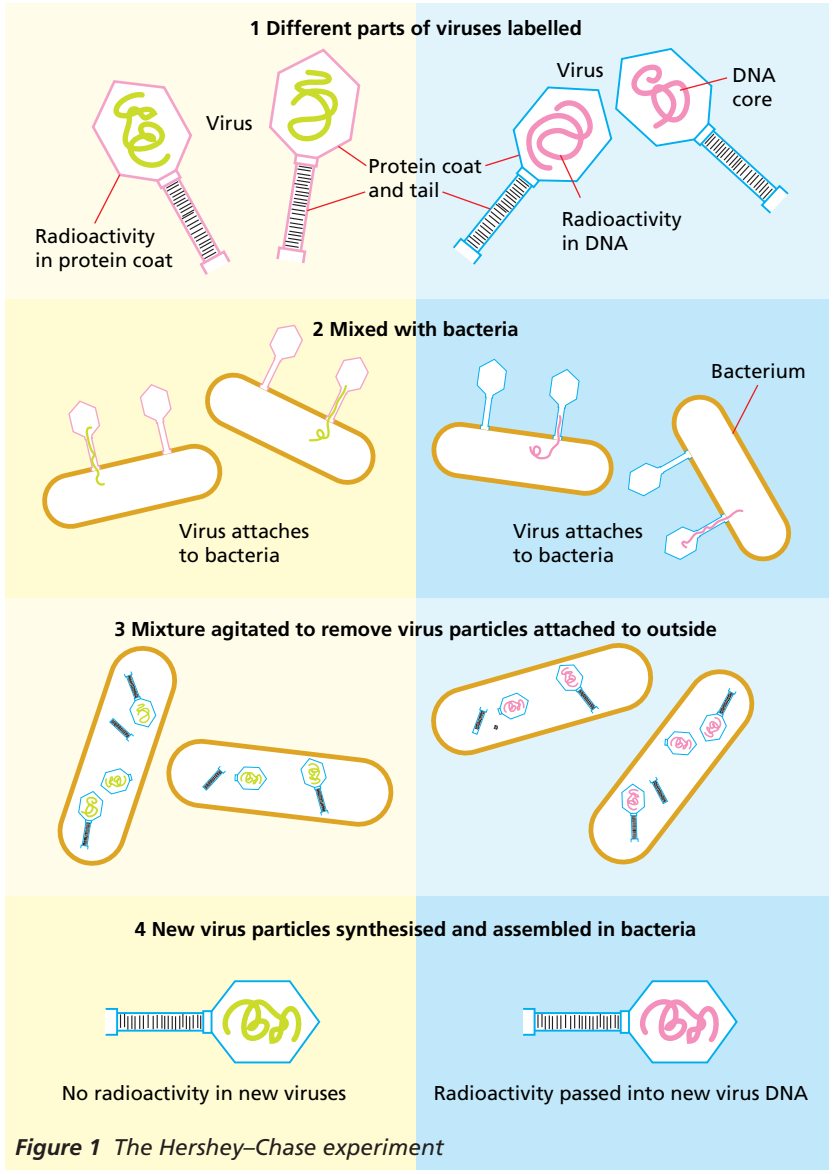


Figure 1 *The Hershey–Chase experiment*

BOX 1 HERSHEY AND CHASE

Hershey and Chase investigated the genetic material of a bacteriophage. Bacteriophages are viruses which attack bacteria and multiply inside them, before killing them. They are very simple in structure, consisting of a protein coat surrounding DNA. Hershey and Chase showed that bacteriophages containing radioactive DNA passed it on to the next generation of bacteriophages. But bacteriophages with radioactive protein did not pass it on to the next generation (see Figure 1).

structure. He noticed that there are the same numbers of molecules of adenine (A) as of thymine (T), and that cytosine (C) matches guanine (G) in the DNA of a given species, although he did not find out why. Many groups of scientists tried to work out the structure of DNA.

WHAT WERE THE VITAL CLUES?

A number of vital clues led the way to understanding the structure of DNA:

- The A–T/C–G base-pair system was deduced.
- In 1948, **Linus Pauling** discovered that many proteins take the shape of an alpha helix, spiralled like a coiled spring.
- **Rosalind Franklin**'s X-ray diffraction pictures of DNA suggested a helix to her and provided several of the vital helical parameters.

On 28 February, 1953, Francis Crick is said to have walked into the Eagle pub in Cambridge and announced, 'We have found the secret of life.' That morning, Watson and Crick had figured out the structure of deoxyribonucleic acid, DNA.

Jane Taylor teaches biology and is an editor of CATALYST.

Answers to wordsquare, page 9

Should genes be patented?

Patenting allows inventors the exclusive right to exploit a product or process they have created — anyone wanting to use the product must pay them a fee. Artificial copies of a gene's DNA sequence can be patented. Many people are worried about the problems this could cause society as a whole. Here Leah Matkin outlines some of the advantages and disadvantages of gene patenting.

For

The economy

Biotechnology industries claim that the right to patent genetic discoveries and products provides the economic incentive for research. Companies seek to recover the huge amonts of money they spend on research by patenting their discoveries. Research companies are rewarded for their discoveries and can use the money gained from patenting to further their research. Human insulin, and enzymes involved in cheese-making, are big earners already.

Social and religious aspects

Knowledge of gene structure has endless beneficial medical uses. For patients with some incurable diseases products developed as a result of gene patenting offer a last hope. Joan Samuelson, who suffers from Parkinson's Disease (a degenerative disease that is incurable but not terminal) states, 'I know that it is our clock that is ticking as we wait for a cure. An ill- thought out scare campaign could stall a breakthrough and doom us to further unnecessary suffering.'

Scientific viewpoints

The development of animals with artificial genes allows important medical research on diseases, and testing of drugs. There is huge potential in the use of artificial DNA sequences in the body as gene therapy to treat inherited conditions such as cystic fibrosis.

Against

The economy

Patenting can discourage new product development because high royalty costs have to be paid to the patent owners. Royalty costs are likely to be passed on to the consumer.

Companies are likely to focus on patenting methods of using the genes they have already patented. This will give them exclusive rights in certain areas, and thus economic advantage. A product resulting from this could be very expensive because it would only be available from one company.

Social and religious aspects

Many religious and animal rights groups believe experiments on genes are inhumane or immoral. The Baptist Convention states, 'We see altering life forms, creating new life forms, as a revolt against the sovereignty of God and an attempt to be God.'

Patents can set dangerous precedents by making living organisms into commodities that can be bought and sold. It also seems highly unfair and unjust that businesses are able to make money from the receptor gene DNA that is found in our own bodies.

Scientific viewpoints

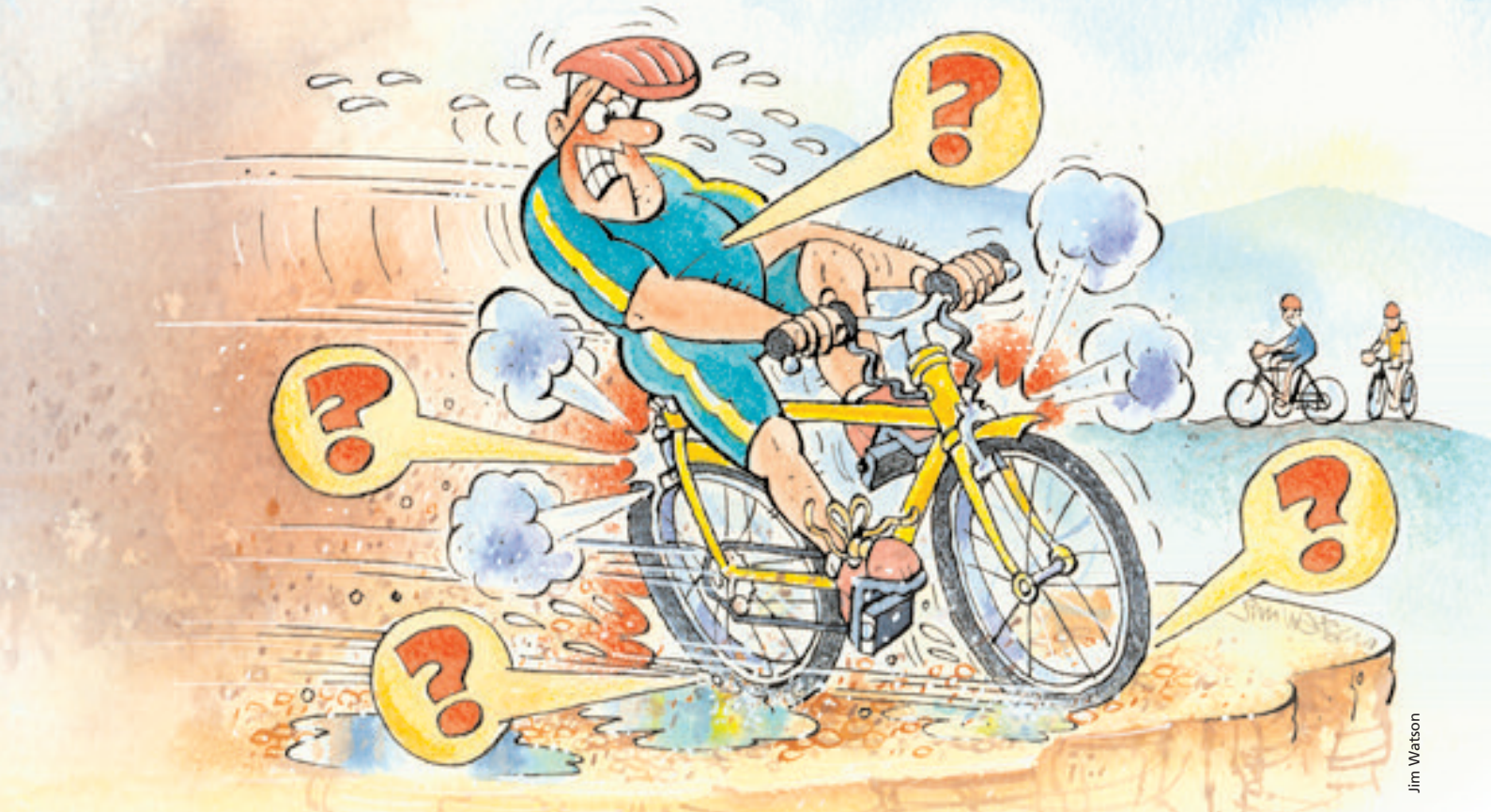
The patenting of living organisms and their parts poses a significant threat to health because restricting access to vital medical and scientific information slows the pace and progress of healthcare. The cost of using patented genes could make research and treatment impossible in developing countries.

Volker Steger/Still Pictures

***Leah Matkin** produced this article as part of her A-level general studies course at Sutton Coldfield GSG. She was exploring the implications for society of scientific advances.*

IMPROVE
your grade

Controlling the variables



Jim Watson

When planning a scientific investigation you need to identify clearly what you are measuring and what may affect the data you record. You will probably vary one factor and see what effect it has on another. Factors that vary are called variables, and you need to know the correct words to describe them, and how to control them.

In real-life situations all sorts of variables operate. In experimental situations you want only a few variables to take into account, or the investigation can become very complicated.

DIFFERENT KINDS OF VARIABLES

The variables that you change deliberately are called **independent variables**. In other words, in the experiment or investigation they are controlled by *you* and do not depend on another variable. Sometimes they are also called **input variables**.

When an input variable is altered it often affects another — the **dependent variable**. In other words this variable depends on another (or maybe on several). It is also sometimes called the **outcome variable**.

Dependent variables may be affected by a number of independent variables at any one time, so in most experiments you need to set things up so that only one independent variable is involved. All others should be kept the same — they are the so-called **controlled variables**. Table 1 provides examples. If you pay attention to controlling the variables you are more likely to make your experiment valid — you will have conducted a 'fair test' and stand a good chance of drawing a valid conclusion.

HOW MANY READINGS?

In many experiments you also need to ensure that you have selected an adequate range of measurements to answer the question you posed. For

Table 1 Variables

Experiment title	Independent variable	Dependent variable	Controlled variables
Rate of reaction of hydrochloric acid with calcium carbonate	Acid concentration	Volume of gas collected per unit time	Temperature Stirring rate Particle size of calcium carbonate
Factors affecting rate of photosynthesis in Canadian pond weed	Light intensity	Volume of oxygen produced, or bubbling rate	Temperature Carbon dioxide concentration
Action of biological and non-biological washing powders	Type of washing powder	Time taken for a stain to disappear	Stain Temperature Amount of powder Volume of water Wash time Amount of stirring
Factors affecting stopping distance of a bicycle	Speed of bicycle	Stopping distance	Braking force Mass of rider Nature of surface
Factors affecting pulse rate	Duration of exercise	Pulse rate	Height of step Steady rate of step ups Recovery between successive readings Emotional state?

The list of controlled variables in Table 1 is not exhaustive. Your knowledge and imagination might allow you to add to them. Note that you can switch controlled and independent variables to extend your investigation of the effects on a particular dependent variable.

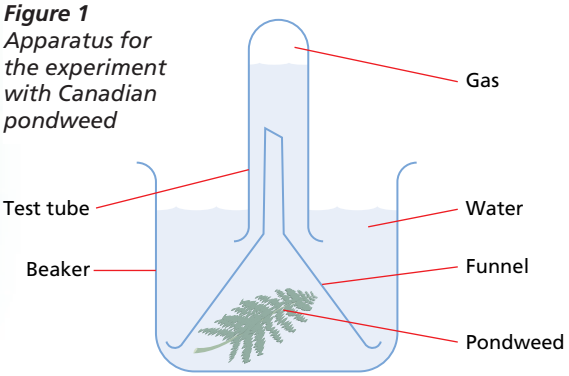


Figure 1 *Apparatus for the experiment with Canadian pondweed*

BOX 1 SOME FURTHER TERMS

You can make observations about variables — things that change or can be changed — in various ways. You might measure and record a simple description of what you see — perhaps a table tennis ball bounces higher than a squash ball when dropped from the same height. This is described as a **categoric variable**. You might count how many times the table tennis ball bounces — this is a **discrete variable**. Because such a variable always involves whole numbers it is sometimes also said to be **discontinuous**. It follows from this that **continuous** variables can have a whole range of numerical values taken anywhere across the range of possible values.

example, you would use quite a wide range of temperatures when assessing the effect of temperature on enzyme activity, not just a few degrees either side of room temperature.

You also need to have an adequate number of readings, both in terms of the intervals across the range and the number of repeats at each interval. A rule of thumb is a range involving five values and three repeats of each. Note that repeating a set of observations three times does not make them more accurate — you might be misreading an instrument or counting or timing inaccurately. But where there should be a pattern in your results, it will help to show up chance errors.

CONTROLS OF ANOTHER KIND

In some experiments you need to adopt a different approach to ensure that a conclusion is valid. You need a **control**, or a series of controls, to rule out other possible explanations.

A simple example relates to a common demonstration — the production of gas by Canadian pondweed. If sprigs of weed are placed under an inverted funnel in a beaker of water with an inverted test tube full of water on top, gas collects in the tube, provided the apparatus is set up in the light (Figure 1). This in itself proves little. You need the same apparatus set up in the dark to show that light is necessary for the gas to be produced.

Is this enough? Suppose you only observed the experiment at the beginning and the end and did not spot the bubbles breaking away from the plant? Could the result be merely a consequence of setting up such a piece of apparatus and exposing water to light?

Perhaps you don't need this control in this experiment but, as with all investigations, it pays to think hard about alternative explanations before you make bold statements about what your results show.

A derived variable is one that is calculated from other variables, which first have to be measured themselves.

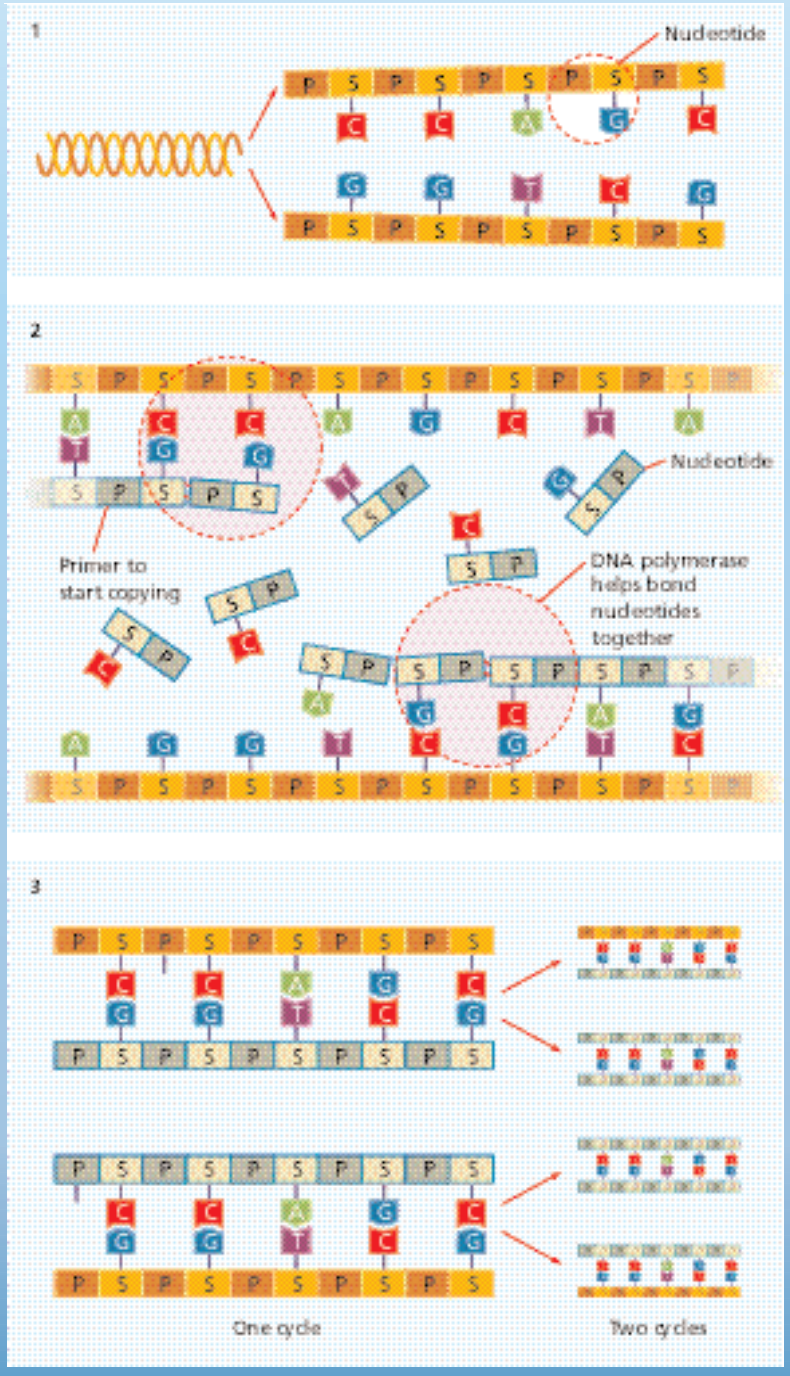
Nigel Collins *teaches biology and is an editor of* CATALYST.

Copying DNA

Many procedures involving DNA require substantial amounts of the chemical. When DNA fingerprinting or profiling is being carried out (see page 2) the samples available, such as those from a crime scene, may be very small, and more may be needed. It is also necessary to increase the amounts of DNA available when working out the sequences of bases. Kary Mullis came up with a way of making multiple copies of DNA molecules which uses the base pairing of the DNA molecule and its ability to copy itself.

- The DNA strands are separated by heating them to 95°C for 5 minutes **(1)**.
- Primers and DNA polymerase enzymes are then added, together with lots of nucleotides. These are units of sugar and phosphate attached to one of the four possible bases. Primers are short lengths of single-stranded DNA. They are made of a sequence of bases put together so that they match the base sequence at the start of a piece of DNA that is of interest.
- The mixture is cooled to 60°C and incubated for a few minutes. During this time primers attach to the single-stranded DNA **(2)**. The enzyme DNA polymerase helps the complementary strand to form.
- Two copies of the molecule are formed after one cycle of heating and cooling **(3)**. About 25 cycles can be fitted into a 2-hour period. This process is known as a polymerase chain reaction (PCR).

Kary Mullis was awarded the Nobel prize for chemistry in 1993 for his development of the polymerase chain reaction 10 years earlier.



WHY ISN'T THE ENZYME DENATURED?

Normally heating an enzyme to 95°C would denature it — DNA polymerase would not survive the heating phase of the second cycle. However, DNA polymerase from bacteria living in hot springs still works at high temperatures. This version of the enzyme is now used in PCR, surviving from cycle to cycle.

WORK IT OUT

After two cycles there are four copies of the DNA, after three there are eight copies and so on.
How many after 10 cycles?
And after 25 cycles? (Answers right)

ANSWERS

After 10 cycles there are 1024 molecules of DNA.
After 25 cycles there are 33 554 432 molecules of DNA.