Cystic fibrosis (CF) is the most common inherited disorder in children of white Caucasian descent, affecting about 1 in 2500 births in the UK. There are about 7,000 CF patients currently registered in the UK. CF is caused by a recessive mutation in a gene which codes for a protein called the cystic fibrosis transmembrane conductance regulator (CFTR) in the plasma membrane of epithelial cells as shown in Fig. 1 below. 1 in 26 of the population are carriers of the disorder.

The CFTR gene codes for a 1480 amino acid protein. It is a large gene and so far more than 800 different mutations have been described for it. Most of these mutations are very rare. The exception is a deletion of 3 nucleotides which codes for a phenylalanine residue at position 508 of the CFTR protein sequence. It is called ΔF508 and accounts for about 70% of CF mutations worldwide. Testing for this mutation and 19 others identifies more than 90% of all mutations. Some mutations result in no CFTR protein being synthesised at all; in others, such as the ΔF508 mutation, the structure of the protein is incomplete and it becomes stuck in the endoplasmic reticulum, never reaching the plasma membrane. In other mutations the protein reaches the membrane but it does not function normally.

The protein coded for by the CFTR gene controls the movement of chloride ions into and out of epithelial cells, such as those lining the airways of the lung. In children born with CF these chloride channels are defective or absent. This results in an abnormal concentration of chloride outside the cell, which in turn causes the mucus that coats epithelial cells to become thicker and stickier than normal.

The mucus builds up in the pancreas, lungs, digestive tract and other organs and can harbour persistent and recurrent infections. Daily physiotherapy, a special diet, antibiotics and other preventative treatments can improve the quality of life for sufferers.

Those suffering from CF may only develop some conditions in adulthood; these include diabetes, cirrhosis of the liver and infertility (especially in men).

Fig 1. Structure of CFTR channel indicating the predicted location of some CF-associated mutations (asterisks). MSD, membrane-spanning domain; NBD, nucleotide-binding domain; R, R domain; Inside, intracellular; and Outside, extracellular. The branched structures are sites where carbohydrate groups are attached. Reproduced with permission from D. Sheppard, Bristol University.
Activity: Answer the following questions:

1. The CF allele is inherited in a typical Mendelian fashion. Carriers of the disease show no symptoms. Using appropriate symbols for the dominant and recessive forms of the CF gene, give the genotypes and phenotypes for:
   a) a CF sufferer;
   b) a CF carrier;
   c) a “normal” individual.

2. Draw a genetic diagram to show the genotypes of children born to parents both of whom are CF carriers. What proportion of the children will be carriers for the disease?

3. 54 million people in the UK are of white European origin. Using the information given above calculate:
   a) the number of CF carriers in the UK;
   b) the chance of two carriers getting married.

4. Boys and girls have an equal chance of getting CF, using all the information you now have, decide which of the following terms best describes CF?
   - sex linked
   - autosomal recessive
   - autosomal dominant

5. a) How many nucleotides code for the amino acids found in the normal CFTR gene?
   b) How many nucleotides code for the amino acids in the CFTR gene in sufferers carrying the ΔF508 mutation?
   c) Describe how the polypeptide chain generated from this faulty gene differs from the normal product.

6. Explain how a couple tested for the ΔF508 mutation could still produce a child with CF.

Extra question:

If you were planning to start a family, would you wish to find out if you and your partner were carriers of CF? How would the results affect your plans?
The most common CF mutations are routinely tested for in many laboratories in the UK. Anyone who has CF in their family is offered a test when planning a family. Both partners need to take the test.

The test involves taking a simple mouthwash. Cells are collected in a weak sugar solution rinsed around the mouth. DNA is extracted from these cells and a diagnostic test using PCR (polymerase chain reaction) is carried out on the DNA samples. If there is a family history for a particular mutation, only that mutation needs to be tested for, but in the partner who does not have a family history up to 20 of the most common gene mutations may be tested for using this technique.

In the tests shown in Fig. 1 below seven individuals have been tested for four different CF mutations. There are two tracks for each DNA sample. ΔF508 and 621+1 show up normally in the first track; G551D and G542X show up normally in track two. A signal in the “wrong” track means that the individual carries the mutation and if the signal is absent in the normal track then the sample is homozygous for the mutation.

![Image of gel with bands indicating presence of mutations](image)

Fig. 1 The results from analysing 7 individuals for the 4 commonest cystic fibrosis mutations in North-West England are shown. The test was developed by Zeneca Diagnostics in collaboration with the Molecular Genetics Laboratory in the Department of Clinical Genetics Royal Manchester Children’s Hospital. Reproduced with permission from Dr. M. Super.

1. Your task is to describe each of the individuals shown in Fig. 1 using the terms normal, heterozygote, homozygote compound heterozygote (when two different mutations are present in the same individual) and the 4 different mutations. The first one has been done for you.
   Individual 1 is NORMAL.
   Individual 2 ..................................................
   Individual 3 ..................................................
   Individual 4 ..................................................
   Individual 5 ..................................................
   Individual 6 ..................................................
   Individual 7 ..................................................

2. Which individuals are
   a) Normal   b) Carriers   c) Affected

3. Using appropriate symbols, draw a pedigree of the possible outcome if Simon (individual 3) and Louise (individual 6) were to produce a family.
ACTIVITY: Should Mollie and Carlos have a baby?

Before starting this activity collect as much information as you can about cystic fibrosis.

Suggested reading:
- Carlos and Mollie’s story Chapter 4 “Your Genes, Your Choices”. In “Exploring the issues raised by genetic research” by Catherine Baker. AAAS. Available on the Internet at http://www.ornl.gov.
- “Genes and You: Teaching about genetics from a human perspective” by Gill Mullinar. Cross-curricular materials for Key stage 4. Published by the Wellcome Trust.
- MRC Research update 3 “Cystic Fibrosis: the quest for a cure”.
- The MRC web site: http://www.hgu.mrc.ac.uk/Research
- Support notes in this pack.
- Class resources

Background
Mollie and Carlos need to decide whether or not to have prenatal testing for cystic fibrosis before trying for a baby. Carlos had a brother who was constantly ill and died young from cystic fibrosis. He has already been tested and knows he is a carrier of the disorder and does not want to see another child suffer as his brother did. He would like Mollie to be tested before they go ahead and start a family. Mollie has no family history of cystic fibrosis and thinks they should take a chance because they would love the baby whether or not it had cystic fibrosis. What should they do?

Activity
There are a number of issues to consider before Mollie and Carlos can decide which path to take. Your task is to consider each of these then decide what you would do if you were Mollie or Carlos.

Use the following questions and ideas to help form your case:
- Find out what you can about CF (cystic fibrosis) – how it is caused; its symptoms; life expectancy of sufferers.
- How common is CF?
- Carlos is a “carrier” for CF – explain.
- Boys and girls have an equal chance of getting CF – what does this tell you about the gene responsible?
- If Mollie decides to be tested for CF, what are the chances they will have an affected baby if a) she is not a carrier? b) she is a carrier?
- If they decide to go ahead and try for a baby, several prenatal tests are available, including alpha-fetoprotein test (AFP), ultrasound, amniocentesis and chorionic villus sampling. Briefly describe each of these and place them in order of risk to the foetus and usefulness (from greatest to least risk).
- Is it possible that Mollie and Carlos could still have a CF baby if the normal tests are negative?
- What are their choices?
- What is the likely outcome if a foetus is found to have 2 mutated CF genes?
- Other choices include pre-implantation diagnosis – what does this entail and what are the advantages and disadvantages?
- What would you do?
ACTIVITY: Mollie and Carlos go for genetic testing

Mollie refused any prenatal testing and the couple decided to go ahead and have a family. Over the next few years they had two normal, healthy sons, Hamish and Gordon. Their third child, Morag, was a daughter with cystic fibrosis. Soon after Morag was born, Carlos and Mollie decided to take out life insurance policies for the whole family. The insurance company insisted that the entire family underwent genetic testing for a range of genetic disorders including CF before offering cover. This time Mollie agreed.

A small sample of blood was taken from each individual in the family. The white blood cells were isolated from the samples and several genetic tests were carried out using microsatellite analysis. The results for one CF test is given below:

<table>
<thead>
<tr>
<th>INDIVIDUAL</th>
<th>MARKER SIZE (bp)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carlos</td>
<td>185/182</td>
</tr>
<tr>
<td>Mollie</td>
<td>185/182</td>
</tr>
<tr>
<td>Hamish</td>
<td>185/182</td>
</tr>
<tr>
<td>Gordon</td>
<td>185</td>
</tr>
<tr>
<td>Morag</td>
<td>182</td>
</tr>
</tbody>
</table>

Use the information above and in the rest of the pack to answer the following questions:

1. Complete the following table using the terms carrier, normal or CF:

<table>
<thead>
<tr>
<th>INDIVIDUAL</th>
<th>MARKER SIZE (bp)</th>
<th>PHENOTYPE</th>
</tr>
</thead>
<tbody>
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<td>Morag</td>
<td>182</td>
<td></td>
</tr>
</tbody>
</table>

2. Why were only white blood cells used for testing?

3. Describe the process used to produce the information in the tables.

4. Is CF a sex linked disorder? How does the information you have been given support your answer?

5. What is Carlos’ reaction likely to be when he is told about his sons’ results?

6. Describe the mutation detected in the CF test used here.

7. The insurance company has also seen the test results. Will the results affect the families wish to take out life insurance?

8. 1 in 26 of the population are known to be carriers of CF.
   (a) What are the chances of two carriers getting married?
   (b) What are the chances of Hamish’s future wife being a CF carrier?
   (c) What proportion of the offspring produced by two carriers will have cystic fibrosis?
   (d) Why is the proportion given in answer (c) only used as a guideline when counselling prospective parents?