Title	**Workshop on genetic disease and gene therapy**
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Target level	KS4 science, GCSE (or A-level)
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NB: All the figures are modifiable and can be provided as a PPT file, so do not hesitate to contact Maggy Fostier to get the files.

Activity	Length	Content included from the GCSE curriculum	Building on the GCSE curriculum
Effect of mutations on proteins	15 mins	Genetic diseases; DNA encodes for proteins **Terminology**: chromosome; gene; allele; proteins; enzymes	Mutations in DNA can result in non- or partially functional proteins **Terminology**: ligand; receptor; protein complex; mutations
Can you predict if this patient has Cystic Fibrosis?	15 mins	Monohybrid inheritance **Terminology**: genotype; phenotype; dominant; recessive; (un)affected ; carrier; cystic fibrosis	Phenotype resulting from the addition of an extra healthy allele (as in gene therapy). **Terminology**: compound heterozygote
Introduction to gene therapy	20 mins	Structure of animal cells **Terminology**: chromosome; gene; allele; nucleus	Gene therapy procedure **Terminology**: Gene expression; target cells; trisomy.

The three activities follow a natural progression but all can be performed independently.

- Activity 1 gets students to draw the effects of mutations on the resulting proteins. Three activity cards illustrate different types of proteins and help define a dominant versus recessive effect.
- Activity 2 gets students to predict based on genotype whether a patient will have cystic fibrosis. The transition to gene therapy is made by asking what would happen if a copy of a healthy allele was added to the genome of each patient.
- Activity 3 introduces the process of gene therapy and gets students to think about why certain aspects of this technique may be performed as it is.

Workshop on genetic disease and gene therapy

Activity 1: Effect of mutations on proteins – 15-20 mins.

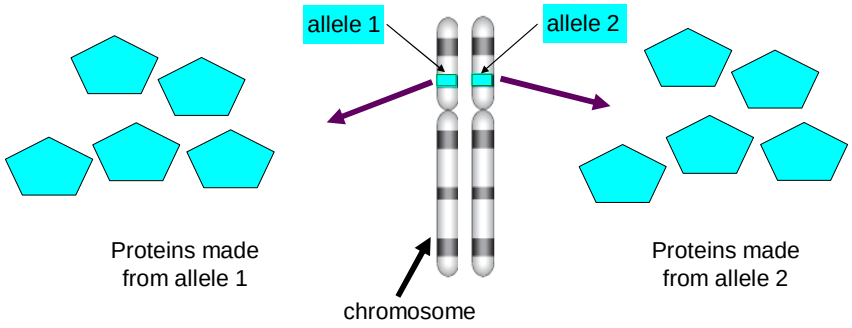
This activity gets students to draw the effects of mutations on the resulting proteins. Three activity cards illustrate different types of proteins and help define a dominant versus recessive effect.

Students work in pairs. The cards below are within a PowerPoint presentation and the slides are modifiable. Each pair should have 1 copy of each slide or the slides could be projected.

The introduction card (if needed) shows that we have two alleles for each gene and each allele produces many copies of the protein it encodes.

Effect of mutations on proteins - Introduction

A gene is a section of DNA that codes for a specific protein. Each gene is made up of two alleles (one on each chromosome), each coding for proteins.



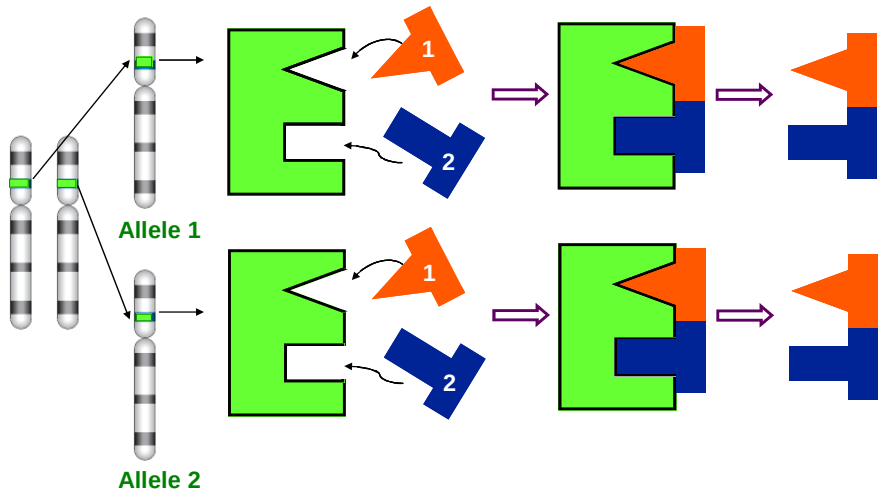
If a mutation occurs in the DNA of an allele, the protein made may have an incorrect structure and not work properly. Alternatively, some mutations can result in no protein being made at all.

The tasks below will make you explore the differences between normal and mutated proteins and how changes in their structure can affect their interactions and function.

Activity card 1: [next page]

Effect of mutations on proteins - Task 1

Some proteins act as an enzyme. Below is an enzyme with two active sites for substrates 1 and 2.



Task: Draw a mutated form of the enzyme for each allele, preventing either substrate to fit into the active sites. The mutations in the two alleles should be the same.

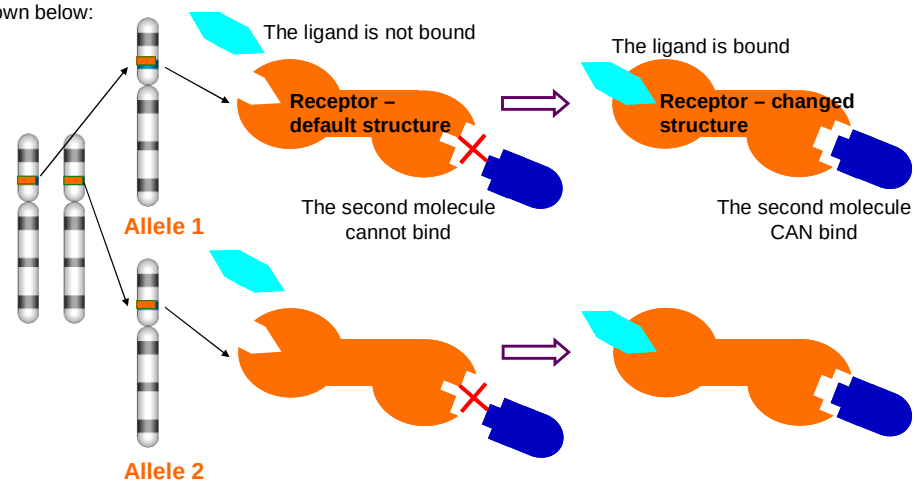
Here students should see that mutations can have multiple effects on proteins and that you need the same mutation on both alleles to get a homozygous effect.

Variant: Get students to draw only one mutated protein. Would it affect the overall function of the cell/body? i.e. Is it likely to result in a recessive or dominant effect? Answer: Probably recessive as the normal enzyme will compensate up to a good level.

Activity card 2:

Effect of mutations on proteins - Task 2

Some proteins act as receptor for other proteins called ligand. After binding with a ligand, the structure of a receptor changes. One result of this could be that a second molecule binds to the receptor as shown below:



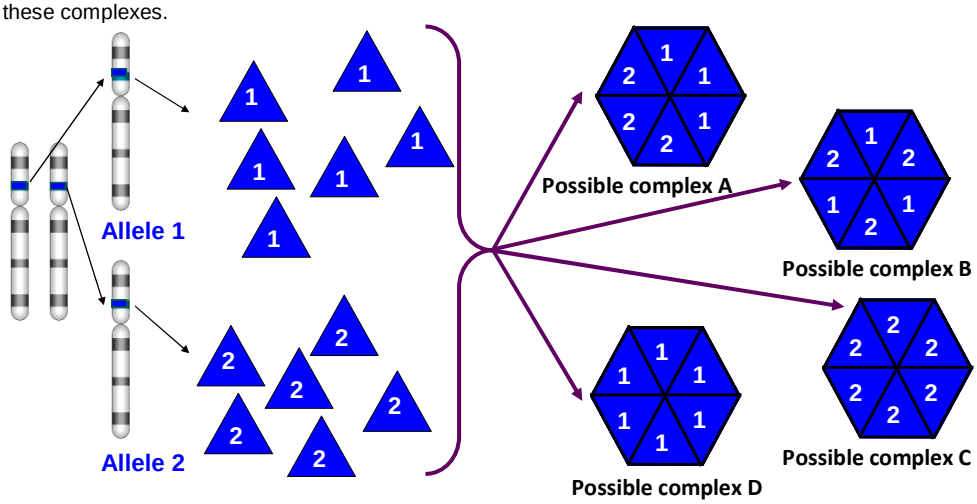
Task: Draw a mutated form of the receptor for each allele, preventing the ligand from binding. The mutations in the two alleles should be different. What happens when the ligand can no longer bind to the receptor?

Here students should appreciate that a person can carry two different mutations (compound heterozygote). This is usually what happens in Cystic Fibrosis
Variant: Get students to draw only one mutated protein. Would it affect the overall function of the cell? i.e. Is it likely to result in a recessive or dominant effect?
Answer: Probably recessive as the normal receptor will compensate up to a good level.

Activity card 3:

Effect of mutations on proteins - Task 3

Some proteins need to form complexes to carry out their function. Below is a complex made by 6 identical proteins interacting with each other. Proteins from allele 1 and 2 assemble randomly into these complexes.



Task: Draw a mutant shape of the protein produced by allele 1 so that it can not longer assemble with the other proteins. Which of the possible complex presented will be affected?

The answer is that all complexes but complex C (only made of proteins from allele 2) will be affected.

Here students should appreciate that this is how a mutation can have a dominant effect. This happens in the disease called Osteogenesis imperfecta or Brittle bone disease, where type I collagen is not produced and so the collagen filaments (protein complexes made of several different proteins) cannot form.

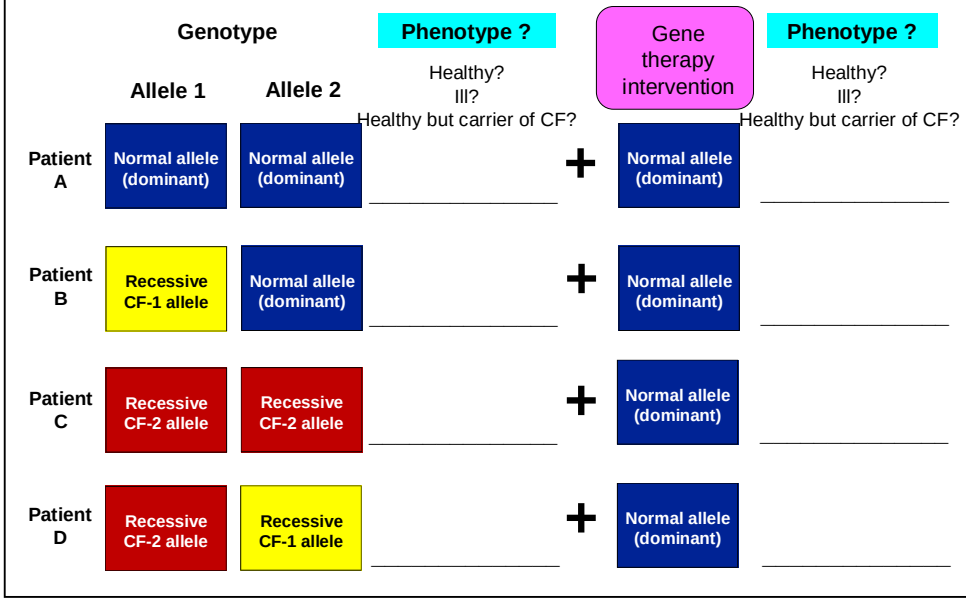
Variant. Instead of drawing the proteins, the students could cut them out (more hands on, but takes longer).

Activity 2: Can you predict if this patient has Cystic Fibrosis? 10 mins

This activity gets students to predict based on genotype whether a patient will have cystic fibrosis. The transition to gene therapy is made by asking what would happen if a copy of a healthy allele was added to the genome of each patient.

Students work in pairs. The card below is within a PowerPoint presentation and the slide is modifiable. Each pair should have 1 copy of the slide or the slide could be projected.

Cystic fibrosis is a disease caused by mutations in a gene coding for a receptor. These mutations are usually recessive. **Task:** Predict the phenotype of each patient.



Patient	Phenotype	Phenotype after gene therapy
A	Healthy	Healthy (although actually having three healthy copies of the gene could lead to over-expression and problems).
B	Healthy but carrier	Healthy but carrier
C	ill	Healthy but carrier **
D	ill *	Healthy but carrier **

** Card 2 of the previous activity can be used to illustrate the types of mutation that the mutations in yellow or red could be.*

*** Adding one extra copy of a normal allele to a patient who carries two recessive mutations is equivalent to cancelling out the effect of one mutation, thus bringing this person back to a normal healthy but carrier status. For a carrier, an extra copy does not change their carrier status as they can still pass on this allele to their progeny.*

NB: The normal allele is neither dominant nor recessive. In combination with a recessive mutation (as here), it is dominant because the normal protein can compensate for the loss of protein function from the mutant. In combination with a dominant mutation, it will behave either as co-dominant or recessive, as the normal protein will only partially compensate or not at all the loss of protein function from the mutant. If you think that adding dominant in the normal allele box may confuse students, then remove it.

This slide illustrates the take home message that gene therapy is the addition of a normal allele to the genome to cancel out the effect of a mutation by adding a dose of normal proteins to the cell – gene therapy is rarely the replacement of a faulty gene.

Activity 3: Introduction to gene therapy. 20 mins

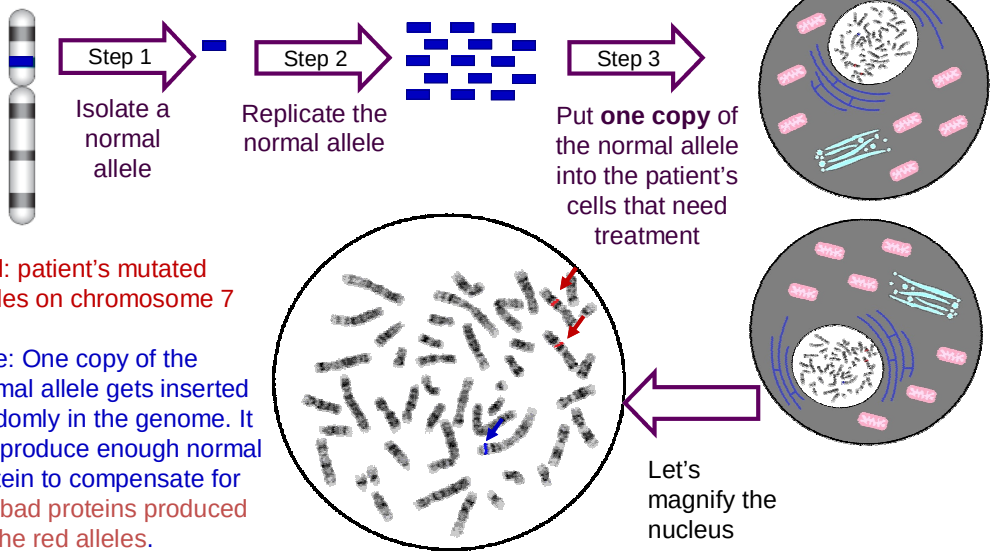
Activity 3 introduces the process of gene therapy and gets students to think about why certain aspects of this technique may be performed as it is.

Students work in pairs. The card below is within a PowerPoint presentation and the slide is modifiable. The worksheet in on the next page

Each pair should have 1 copy of the slide or the slide could be projected.

Effect of gene therapy on a patient suffering from Cystic Fibrosis.

Our patient is affected by CF because he has two copies of a recessive mutation (red below). The gene therapy process consists of adding a copy of the normal allele into the genome of the cells that need it for their function (e.g. cells from the lungs).



Worksheet for gene therapy.

NB: Question 3, 4 and 6 are very advanced; just do your best.

Q1: Is the disorder being treated here dominant / recessive? ……

Q2: This patient has two mutated, disease–causing alleles. In step 1, we isolate the normal allele. Where do you think we could we isolate the normal allele from in order to treat him? ……

Q3: During gene therapy, we insert one copy of a normal allele per cell that needs it. Why don't we insert a whole chromosome from a healthy patient instead?

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Q4: Why is the normal allele not inserted into every cell of the patient (e.g. for CF we aim at the lungs and not the brain nor the heart)?

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Q5: Where does the copy of the normal allele inserts in the cell nucleus?

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Q6: This patient has two mutated allele in its genome, why do we insert only one copy of the normal allele in the cells that need, instead of 2 copies?

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Q7: Select the answer that best summarises the diagram on the card (A, B, C, D, E, or F).

		...in all cells.	...in target cells.
Gene therapy is...	...replacing the mutated alleles...	A	D
	...inserting a normal allele...	B	E
	...repairing the mutated alleles...	C	F

Worksheet for gene therapy. Answers.

Q1: Is the disorder being treated here dominant / recessive? ***Recessive.***

Q2: This patient has two mutated, disease–causing alleles. In step 1, we isolate the normal allele. Where do you think we could we isolate the normal allele from in order to treat him? ***Cells from a person who does not have the disorder.***

Q3: During gene therapy, we insert one copy of a normal allele per cell that needs it. Why don't we insert a whole chromosome from a healthy patient instead?

The normal chromosome would be too big and complicated to insert, but more worryingly, 2) it would cause these cells to have a trisomy of chromosome 7, i.e. three copies of chromosome 7, i.e. each gene on chromosome 7 would have 3 alleles (instead of 2), each producing proteins. This is a problem because our body is designed for the amount of proteins produced by 2 alleles, and in the same way that having too little protein produced is problematic, having too much protein is not good either.
Trisomy is also the term for genetic conditions that can occur when the gametes are being formed. It is usually lethal before birth or causes huge problems in development (e.g. trisomy 13 or 21).

Q4: Why is the normal allele not inserted into every cell of the patient (e.g. for CF we aim at the lungs and not the brain nor the heart)?

Only few genes are required in every cell or our body. Most genes are only expressed as proteins in a subset of cells, e.g. the CF receptor is expressed and used in the lungs and not the brain.
This is actually good news for gene therapy because we would not know how to insert the normal allele in all cells.

Q5: Where does the copy of the normal allele inserts in the cell nucleus?
Randomly on any chromosome.

Q6: This patient has two mutated allele in its genome, why do we insert only one copy of the normal allele in the cells that need, instead of 2 copies?

The main reason is that this random integration of the extra allele on any chromosome can cause mutations/problems in the neighbouring genes of the insertion sites, so inserting one copy is already risky. One copy is also enough as it will produce enough protein to compensate for the loss or not functional proteins in the target cells.

Q7: Select the answer that best summarises the diagram on the card (A, B, C, D, E, or F).

		...in all cells.	...in target cells.
Gene therapy is...	...replacing the mutated alleles...	A	D
	...inserting a normal allele...	B	E
	...repairing the mutated alleles...	C	F

Answer: E

But note that scientists also try to conduct gene therapy with method D and F, as explained on this concise and accurate webpage:
http://www.news-medical.net/health/What-is-Gene-Therapy.aspx