Edexcel

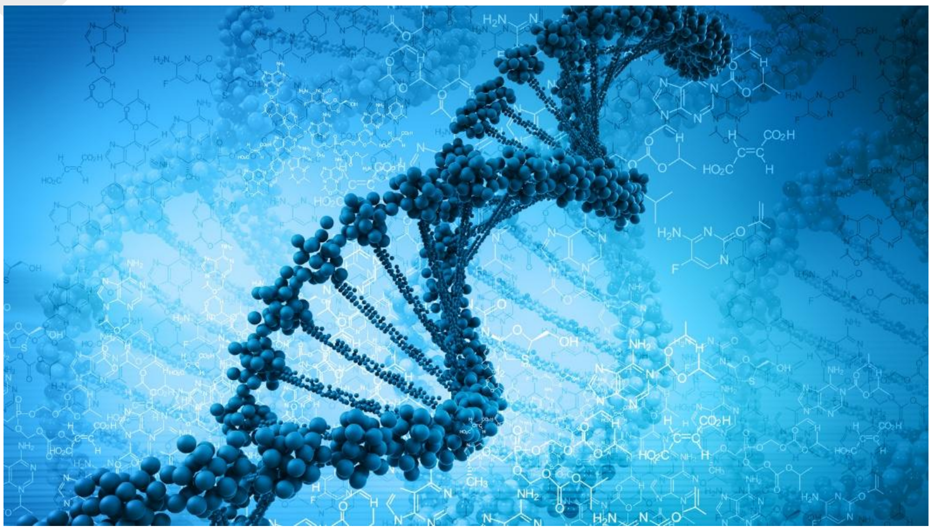
IGCSE

Biology

Unit 05

Code (4BI1)

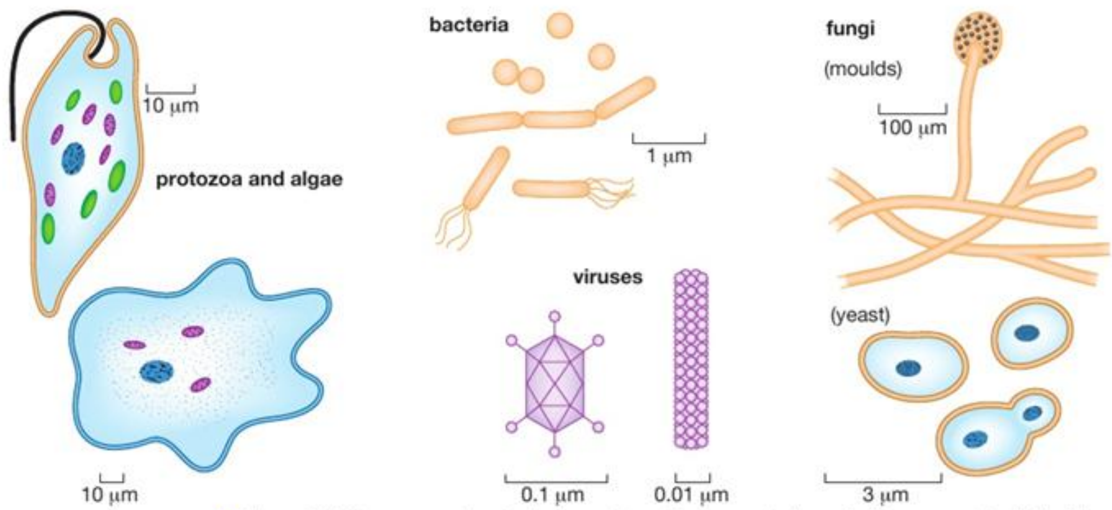
Microorganisms and genetic modification



What is microorganisms?

Microorganisms are living organisms visible only under a microscope. They play a crucial role in recycling waste products and regenerating organisms after death. They cause disease in animals and plants, but humans use their reproductive capacity for food, drink, and medicines. Microorganisms include protoctists, bacteria, viruses, and some fungi.

Figure 21.1 shows a few examples of the many types of microorganisms.



▲ Figure 21.1 Some examples of microorganisms. They are not drawn to the same scale. Notice the range of size, as shown by the scale bar alongside each organism. One micrometre (1 µm) is a millionth of a metre, or a thousandth of a millimetre.

FERMENTATION AND BIOTECHNOLOGY

Fermentation and fermenters are terms that you will encounter if you are involved in the growth of microorganisms.

This overlaps with the definition of **biotechnology**, which means using any organisms (but mainly microbes) to make products that are useful to humans. Although the word itself is relatively new, humans have been using biotechnology processes for thousands of years without knowing it. Since ancient times, fermentation by yeast has been used to produce bread. Yoghurt is made by the action of bacteria on milk, and other bacteria and moulds are used in cheese manufacture.

Modern biotechnology also allows us to alter the genes of microorganisms so that they code for new products. This is called **genetic engineering**. It is a topic that you will read about in Chapter 22.

TRADITIONAL BIOTECHNOLOGY - MAKING FOOD AND DRINKS

When yeast cells are deprived of oxygen, they respire anaerobically, breaking sugar down into ethanol and carbon dioxide:

glucose → ethanol + carbon dioxide

This process has been used for thousands of years to make bread, using a species of yeast called Saccharomyces cerevisiae.

Making drinks

Wine is made using yeast to ferment sugars in grape juice, with commercial production in vats and homemade in small-scale fermenters with an 'airlock'. Alcohol concentration increases until yeast cells die. Beer is made from barley seeds, which are germinated and produce amylase, breaking down starch into sugar maltose. Maltose is fermented by yeast in a large open vat.

Figure 21.2 A small fermenter for producing homemade wine. The U-shaped tube at the top is a water-filled airlock that prevents oxygen entering the fermenter but allows carbon dioxide to escape.

Figure 21.3 A froth forming on the surface of the beer as yeast ferments the sugar to alcohol and carbon dioxide. The carbon dioxide in the froth prevents oxygen entering the mixture from the air – keeping conditions anaerobic.

Making bread

Yeast is also used to make bread. Wheat flour and water are mixed together and yeast added, forming the bread dough. Enzymes from the original cereal grains break down starch to sugars, which are respired by the yeast. Extra sugar may be added at this stage..

Later, when the dough is baked in the oven, the gas bubbles expand. This gives the bread a light, cellular texture (Figure 21.4). Baking also kills the yeast cells and evaporates any ethanol from the fermentation.

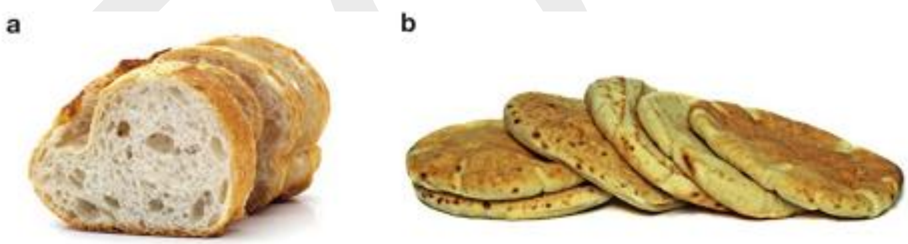


Figure 21.4 (a) The 'holes' in this bread were produced by bubbles of carbon dioxide released from the respiration of the yeast. (b) Bread that is made without yeast is called unleavened bread. What is the difference in texture and appearance between leavened and unleavened bread?

Making yogurt

Yoghurt is milk that has been fermented by certain species of bacteria, called lactic acid bacteria. The effect of the fermentation is to turn the liquid milk into a semi-solid food with a sour taste.

To make yoghurt, milk is first pasteurised at 85-95 °C for 15-30 minutes, to kill any natural bacteria that it contains, then homogenised to disperse the fat globules. The milk is then cooled to 40-45 °C and inoculated with a starter culture of two species of bacteria, called Lactobacillus and Streptococcus.

The drop in pH (as the yoghurt forms) gradually reduces the reproduction of the lactic acid bacteria (although it doesn't kill them). It also helps to prevent the growth of other microorganisms, and so preserves the nutrients in the milk. The steps in yoghurt production are summarised in the flow chart (Figure 21.6).

Milk is pasteurised at 85–95 °C for 15–30 minutes.

↓

Milk is homogenised.

↓

Milk is cooled to 40–45 °C and inoculated with a starter culture of lactic acid bacteria.

↓

Mixture is incubated at this temperature for several hours, while bacteria digest milk proteins and ferment lactose to lactic acid.

↓

Thickened yoghurt is stirred and cooled to 5 °C.

↓

Flavourings, colorants and fruit may be added before packaging.

Figure 21.6 Flow chart showing the stages in yoghurt production.

Industrial fermenters

A fermenter is any vessel that is used to grow microorganisms used for fermentation. Even a baking tray containing a ball of dough could be defined as a fermenter!

Industrial fermenters are large tanks that can hold up to 200 000 dm3 of a liquid culture (Figure 21.7). They enable the environmental conditions such as temperature, oxygen and carbon dioxide concentrations, pH and nutrient supply to be carefully controlled so that the microorganisms will yield their product most efficiently. A simplified diagram of the inside of a fermenter is shown in Figure 21.8.

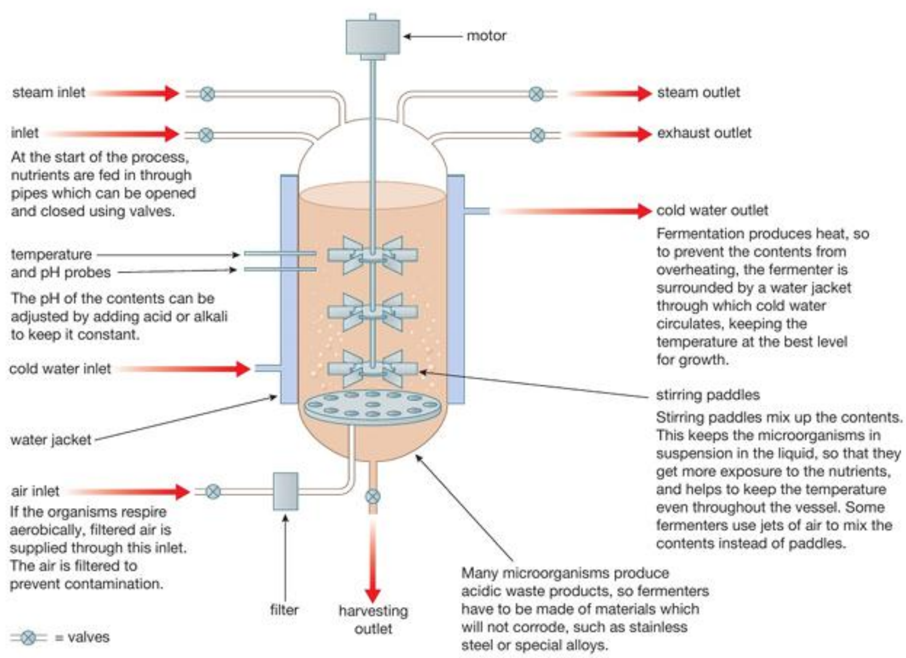


Figure 21.8 An industrial fermenter. Fermenters like this are used to make many products, such as the antibiotic penicillin.

Chapter 22 – genetic modification

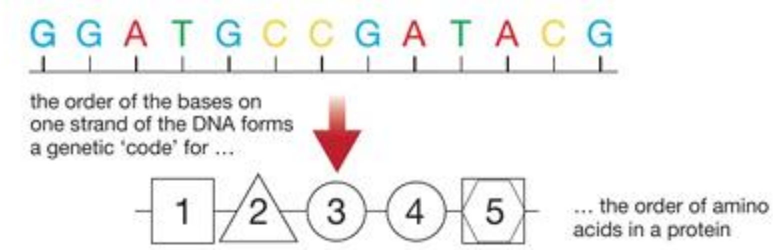


Figure 22.1 The role of DNA in protein synthesis

The protein that is produced could be:

- an enzyme that controls a particular reaction inside a cell or in the digestive system
- a structural protein like keratin in hair, collagen in skin or one of the many proteins found in the membranes of cells
- a protein hormone such as insulin
- a protein with a specific function such as haemoglobin or an antibody.

RECOMBINANT DNA

The production of **recombinant DNA** is the basis of genetic engineering. A section of DNA-a gene- is cut out of the DNA of one species and inserted into the DNA of another. This new DNA is called 'recombinant' DNA because the DNA from two different organisms has been 'recombined'. The organism that receives the gene from a different species is a **transgenic organism.**

Producing genetically modified (transgenic bacteria)

The breakthrough in being able to transfer DNA from cell to cell came when it was found that bacteria have two sorts of DNA-the DNA found in their bacterial 'chromosome' and much smaller circular pieces of DNA called **plasmids** (see Unit 1, Figure 2.10).

■**Restriction endonucleases** (usually shortened to restriction enzymes) are enzymes that cut DNA molecules at specific points. Different restriction enzymes cut DNA at different places. They can be used to cut out specific genes from a molecule of DNA.

■**Ligases** (or DNA ligases) are enzymes that join the cut ends of DNA molecules.

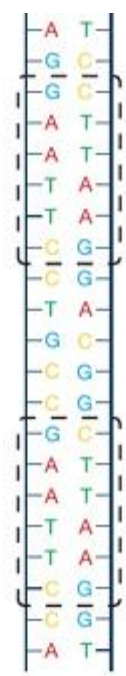
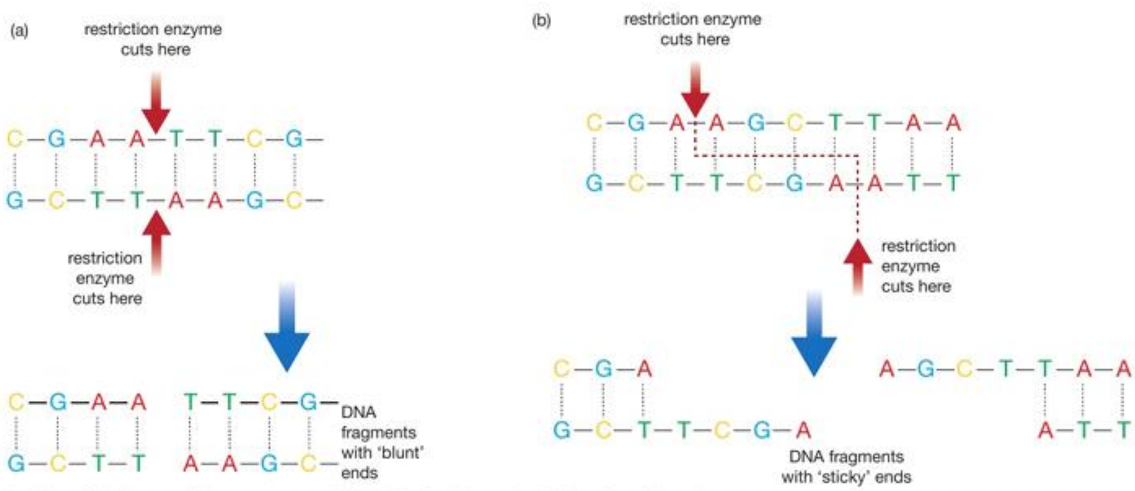
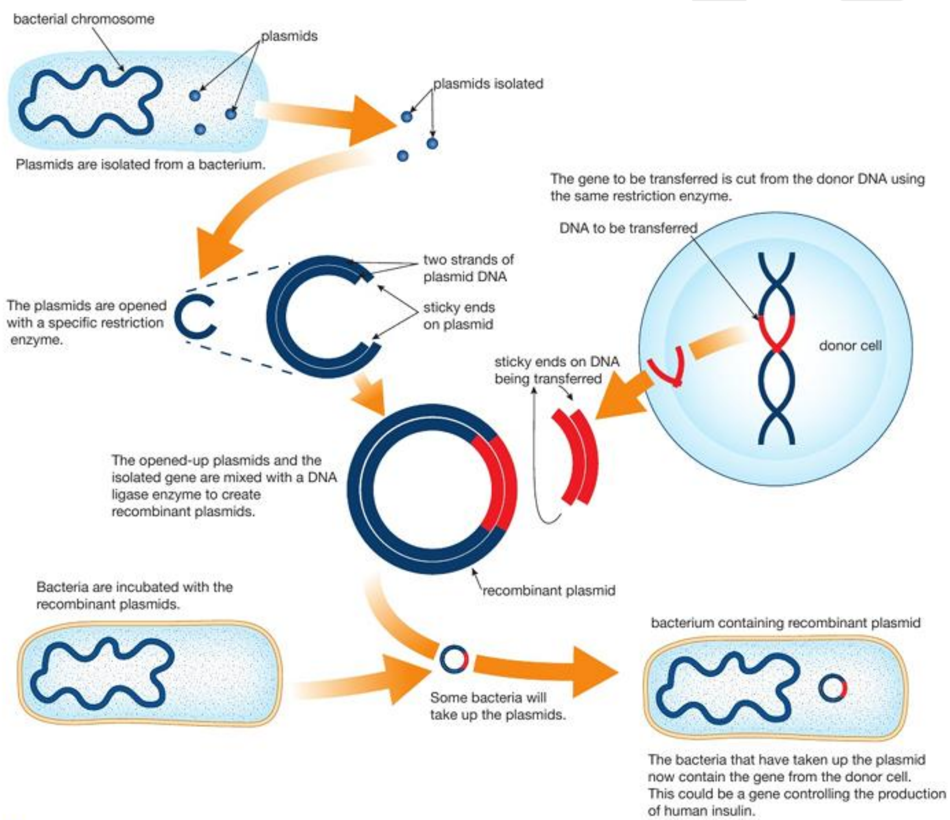


Figure 22.2 Part of a DNA molecule containing the base sequence G-A-A-T-T-C. Notice that the sequence is present on both strands, but running in opposite directions.



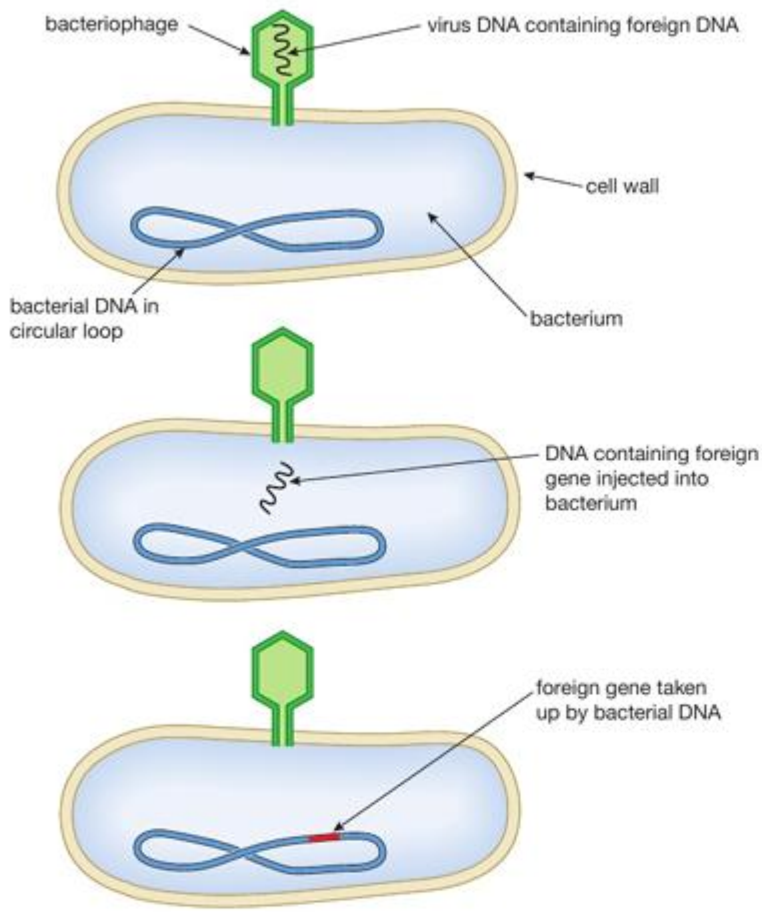
▲ Figure 22.3 How restriction enzymes cut DNA. (a) Forming blunt ends. (b) Forming sticky ends.



▲ Figure 22.4 Stages in producing a transgenic bacterium.

Another vector that has been used to introduce foreign DNA into bacterial cells is the **bacteriophage**. A bacteriophage, or 'phage', is a virus that attacks a bacterium. It does this by attaching to the cell wall of the bacterium and injecting its own DNA into the bacterial cell (Figure 22.5). This DNA becomes incorporated into the DNA of the host cell, and eventually causes the production of many virus particles.

If a foreign gene can be inserted into the DNA of the virus, the virus will inject it into the bacterium along with its own genes. Viruses were used as vectors in the early days of genetic modification, but most gene transfer is now carried out using plasmids.



▲ Figure 22.5 A bacteriophage attacking a bacterial cell.

Making use of genetically bacteria

Different bacteria have been genetically modified to manufacture a range of products. Once they have been genetically modified, they are cultured in fermenters to produce large amounts of the product (see Chapter 21). Some examples are described below.

1. Human insulin People suffering from diabetes need a reliable source of insulin. Before the use of genetic engineering to make human insulin, the only insulin available was extracted from the pancreases of other animals such as cattle. This is not quite the same as human insulin and does not give the same level of control of blood glucose levels.

2. Enzymes for washing powders Many stains on clothing are biological. Blood stains are largely proteins, grease marks are largely lipids. Enzymes can digest these large, insoluble molecules into smaller, soluble ones. These then dissolve in the water. Amylases digest starch, proteases digest proteins and lipases digest lipids. Bacteria have been genetically engineered to produce enzymes that work at higher temperatures, allowing even faster and more effective action.

3. Enzymes in the food industry One bacterial enzyme used in the food industry is glucose isomerase. This enzyme catalyses a reaction which converts glucose into a similar sugar called fructose. Fructose is much sweeter than glucose and so less is needed to sweeten foods. This has two advantages - it saves money (since less is used) and it means that the food contains less sugar and is healthier.

4. Human growth hormone The pituitary gland of some children does not produce enough of this hormone and they show a slow rate of growth. Injections of growth hormone from genetically modified bacteria restore normal growth patterns.

5. Bovine somatotrophin (BST) (a growth hormone in cattle) This hormone increases the milk yield of cows and increases the muscle (meat) production of bulls. Giving injections of BST to dairy cattle can increase the milk yield by up to 10 kg per day. To do this they need more food, but this increased cost is more than offset by the increased income from the increased milk yield (Table 22.1).

6. Human vaccines Bacteria have been genetically modified to produce the antigens of the hepatitis B virus. This is used in the vaccine against hepatitis B. The body makes antibodies against the antigens but there is no risk of contracting the actual disease from the vaccination.

Table 22.1 Effects of BST on milk yield.

	Feed / kg per day	Milk output / kg per day	Milk to feed ratio
without BST	34.1	27.9	0.82
with BST	37.8	37.3	0.99

PRODUCING GENETICALLY MODIFIED PLANTS

The gene technology described so far can transfer DNA from one cell to another cell. In the case of bacteria, this is fine - a bacterium only has one cell. But plants have billions of cells and to genetically modify a plant, each cell must receive the new gene. So, any procedure for genetically modifying plants has two main stages:

- introducing the new gene or genes into plant cells
- producing whole plants from just a few cells.

Biologists initially had problems in inserting genes into plant cells. They then discovered a soil bacterium called Agrobacterium, which regularly inserts plasmids into plant cells. Now that a vector had been found, the rest became possible. Figure 22.6 outlines one procedure that uses Agrobacterium as a vector.

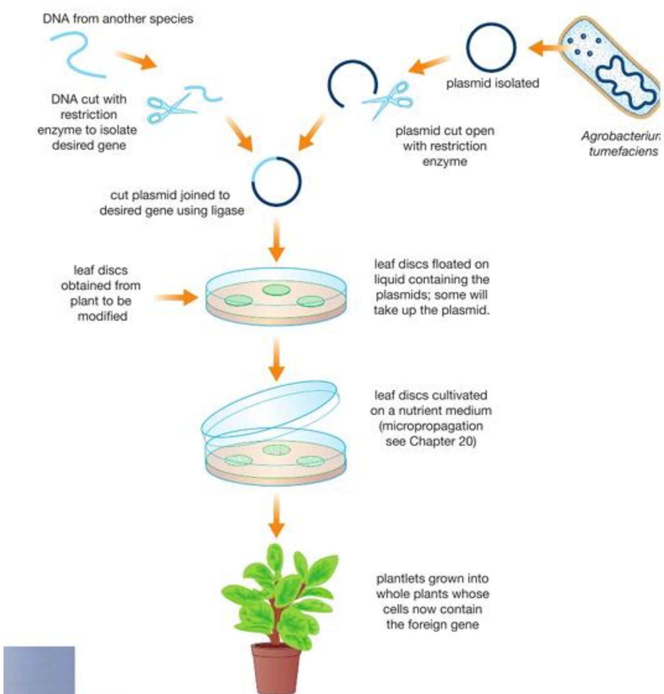


Figure 22.6 Genetically modifying plants using *Agrobacterium*.

Making use of genetically plants

Genetically modified plants are already available to growers and farmers, with some being engineered for extended shelf lives and herbicide resistance. Some plants have been genetically modified to be frost-resistant, while others have been genetically modified to produce beta-carotene, a chemical that gives carrots their color and color to rice. This could potentially save the eyesight of millions of children in less economically developed countries who lack enough vitamin A in their diet.

▲ Figure 22.7 A gene gun

▲ Figure 22.8 Golden rice

Besides the specific examples given, research into the genetic modification of plants provides, or hopes to provide, plants with:

- increased resistance to a range of pests and pathogens
- increased heat and drought tolerance
- increased salt tolerance
- a better balance of proteins, carbohydrates, lipids, vitamins and minerals.

PRODUCING GENETICALLY MODIFIED ANIMALS

Scientists researching the production of genetically modified animals had to find other techniques. The most successful involves injecting DNA directly into a newly fertilised egg cell. This develops into an embryo, then an adult (Figure 22.9).

Research of this kind can produce beneficial results similar to those achieved by genetically modifying plants, such as:

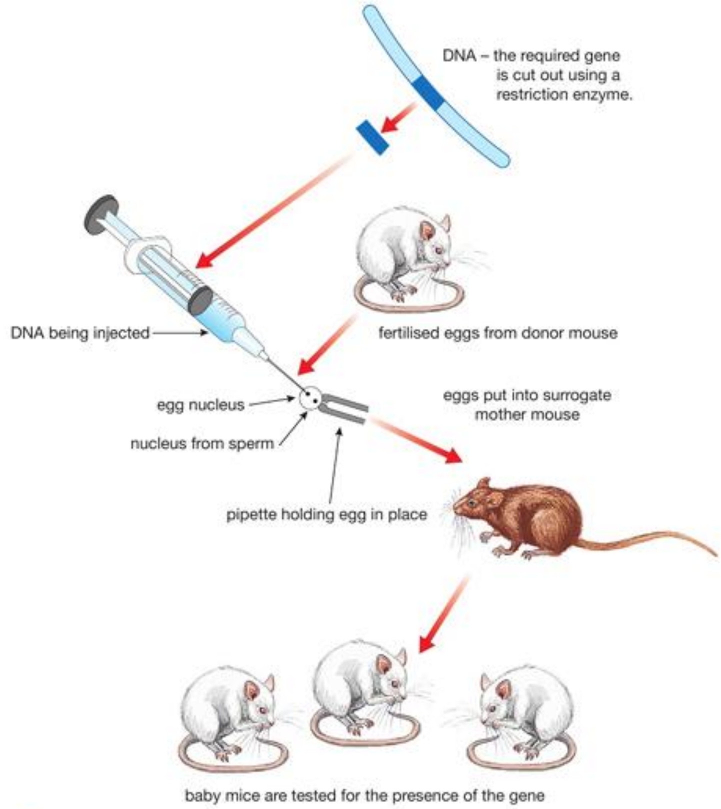
■manufacture of human proteins, such as antibodies, blood clotting factors or alpha-1-antitrypsin (AAT) - see below

■increased production of a particular product, e.g. higher milk yield in cows, greater muscle mass in animals used for meat increased resistance to disease

■production of organs for transplantation (xenotransplantation).

cloning trans genetic animals

Cloning genetically modified animals could produce large, identical populations for the production of human proteins like alpha-1-antitrypsin (AAT), which is involved in the immune response. Some people carry mutations that increase AAT production, potentially treating those with AAT deficiency. Cloning transgenic sheep could increase the amount of AAT available.



▲ Figure 22.9 The procedures used in producing genetically modified animals.

Revision questions

1. Plants can be genetically modified (GM) to make them resistant to pests. Describe an investigation that could be carried out to find out if GM plants produce a better yield than normal plants. Your answer should include experimental details and be written in full sentences.

2. Human insulin can be made by genetically modified bacteria.

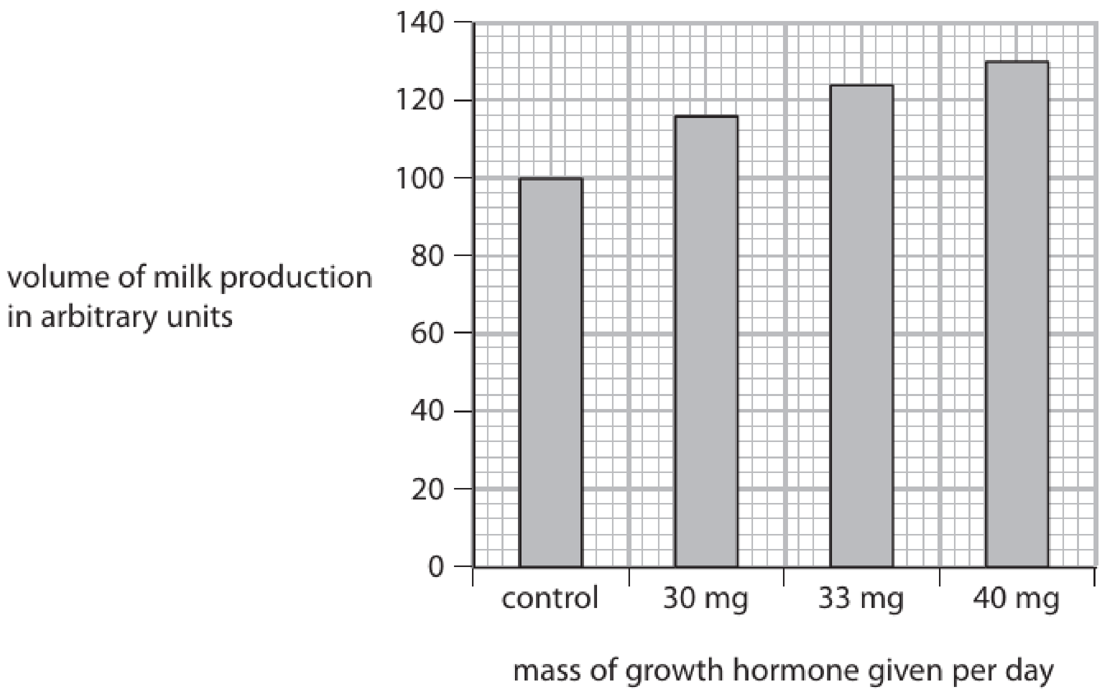
(a) (i) Name the small circle of DNA that is genetically modified in bacteria.

(ii) Name two enzymes that are used to genetically modify the DNA of the bacteria.

(b) To produce large amounts of human insulin the genetically modified bacteria are grown in a fermenter. Describe an investigation to find out if temperature affects the amount of insulin made by genetically modified bacteria. Your answer should include experimental details and be written in full sentences and paragraphs.

3. Describe the stages by which a bacterium can be genetically modified to produce large amounts of a named human protein.

4. An investigation was carried out to find out the effect of a growth hormone on milk production. Groups of cows were given different masses of a growth hormone. The volume of milk the cows produced was then measured. The graph shows the results.



(a) (i) How much growth hormone should have been given to the control group?

(ii) Describe the effect of growth hormone on milk production.

(b) Farmers want to make reliable comparisons about the effect of different doses of growth hormone.

(i) What was done in this investigation to make the results reliable?

(ii)Many variables that affect milk production need to be kept the same for each group of cows. This allows a valid comparison to be made between each group. Give two variables that need to be kept the same.

(c) Growth hormone is a protein. It might be present in the milk produced by the cows and then be consumed by humans. Some people are worried that this may harm humans. Other people say that this is not a problem for two reasons. Firstly, the milk is pasteurised (heated to high temperatures). Secondly, the growth hormone is destroyed in the human stomach.

(i) Suggest what happens to the growth hormone when milk is pasteurised.

(ii) Describe how the growth hormone could be destroyed in the stomach

(d) The growth hormone used in this investigation was obtained from genetically modified bacteria. Describe how bacteria can be genetically modified and used to produce growth hormone.

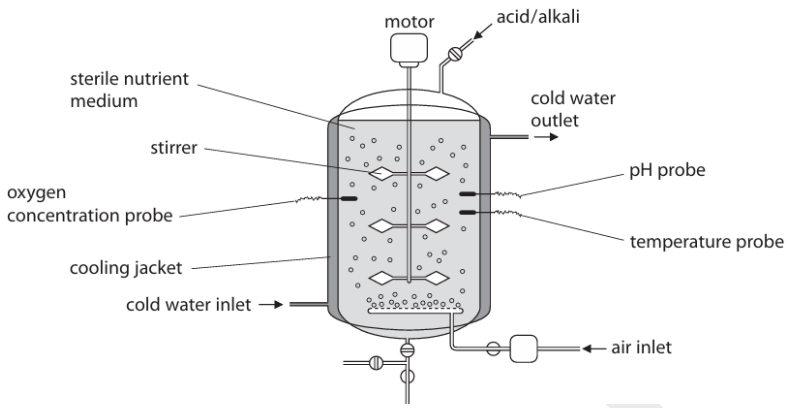
5. Insulin is an important hormone.

(a) Name the organ that produces insulin

(b) State the role of insulin in the body.

(c) (i) Describe how bacteria can be genetically modified to produce human insulin.

(d) The diagram shows part of a fermenter used to grow large numbers of genetically modified bacteria.



(i) Suggest how the air inlet helps the genetically modified bacteria to grow.

(ii) If the pH probe stops working the pH in the fermenter becomes more acidic. Describe and explain how this affects the production of human insulin

6. The picture shows a sheep that has been genetically modified to contain a human gene for making a human protein in its milk

The protein in its milk is a blood clotting substance called factor IX.

(a) The process of genetic modification used to produce this sheep involves the use of two types of enzyme. One enzyme cuts DNA and the other enzyme joins DNA. The process also used a vector.

(i) Name the enzyme that cuts DNA.

(ii) Name the enzyme that joins DNA.

(iii)Name a vector

(b) This sheep is transgenic. What is meant by the term transgenic?

(c) The transgenic sheep can be reproduced by cloning. Suggest the advantages of reproducing the transgenic sheep by cloning.

d) (i) Name the small structures in normal plasma that are involved in blood clotting.

7. Plants can be genetically modified (GM) to make them resistant to pests. Describe an investigation that could be carried out to find out if GM plants produce a better yield than normal plants. Your answer should include experimental details and be written in full sentences

8. Human insulin can be made by genetically modified bacteria.

(a) (i) Name the small circle of DNA that is genetically modified in bacteria.

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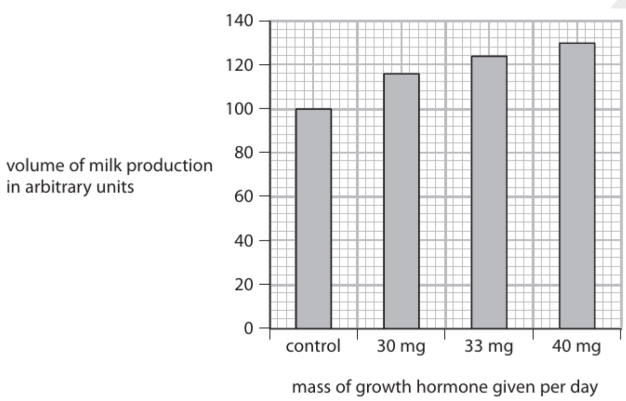
9. Describe the stages by which a bacterium can be genetically modified to produce large amounts of a named human protein.

10. An investigation was carried out to find out the effect of a growth hormone on milk production. Groups of cows were given different masses of a growth hormone. The volume of milk the cows produced was then measured. The graph shows the results.



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d) The growth hormone used in this investigation was obtained from genetically modified bacteria. Describe how bacteria can be genetically modified and used to produce growth hormone.