Chapter 20 – Molecular Genetics

Subject content

Content
- The Structure of DNA
- The Role of DNA in Protein Synthesis
- Genes
- Genetic Engineering and Medical Biotechnology

Learning outcomes
(a) outline the relationship between DNA, genes and chromosomes
(b) state the structure of DNA in terms of the bases, sugar and phosphate groups found in each of their nucleotides
(c) state the rule of complementary base pairing
(d) state that DNA is used to carry the genetic code, which is used to synthesise specific polypeptides (details of transcription and translation are not required)
(e) state that each gene is a sequence of nucleotides, as part of a DNA molecule
(f) explain that genes may be transferred between cells. Reference should be made to the transfer of genes between organisms of the same species or different species – transgenic plants or animals
(g) briefly explain how a gene that controls the production of human insulin can be inserted into bacterial DNA to produce human insulin in medical biotechnology
(h) discuss the social and ethical implications of genetic engineering, with reference to a named example

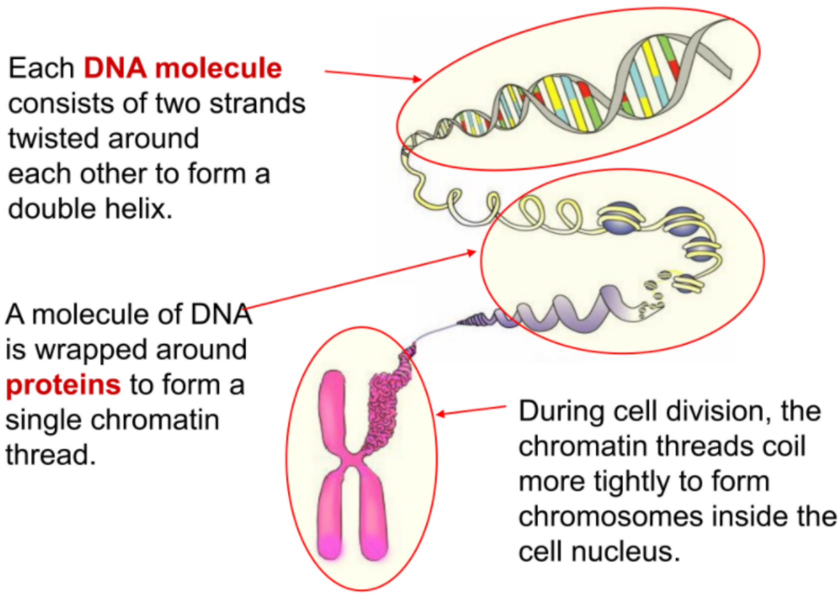
Use the knowledge gained in this section in new situations or to solve related problems.

Definition

Phrase	Definition
Deoxyribonucleic acid (DNA)	Molecule that carries genetic information (important for all cellular functions – cell division, cell differentiation)
Chromosome	DNA molecule coil around histone proteins to form chromatin thread During cell division, chromatin thread condenses to form chromosome
Gene	A unit of inheritance borne on a particular locus of a chromosome A sequence of DNA nucleotides that code for a polypeptide to make a protein
Genetic code	Message stored by gene that determines how a protein should be made in a cell
Transcription	Process where DNA template is used to make a messenger RNA (mRNA) molecule
Translation	Process where sequence of mRNA codons is used to make polypeptide

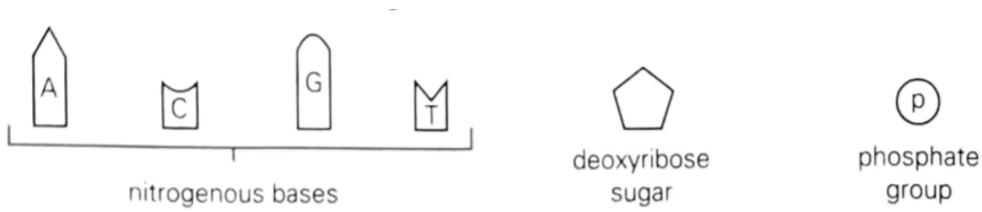
Genetic engineering	Technique used to transfer genes from one organism to another. Individual genes are cut off from cells of one organism & inserted into cells of another organism of same / different species. Transferred gene can express itself in recipient organism.
Transgenic organism	Organism which acquires foreign gene

20.1 DNA
<u>Organisation of DNA in cells</u>

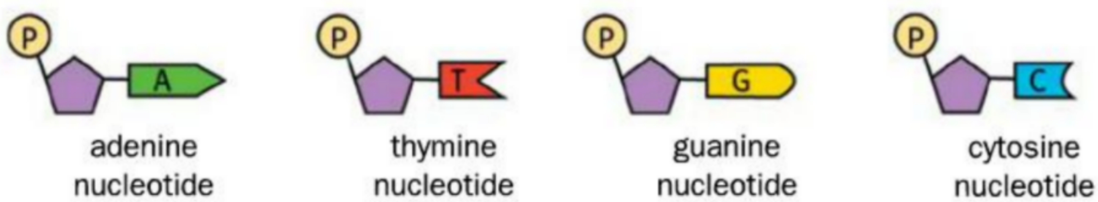


Complexity
1. **Gene**: segment of DNA
2. **DNA molecule**
3. **Chromatin thread**: DNA coiled around protein molecules
4. **Chromosome**: chromatin thread condense (cell division)

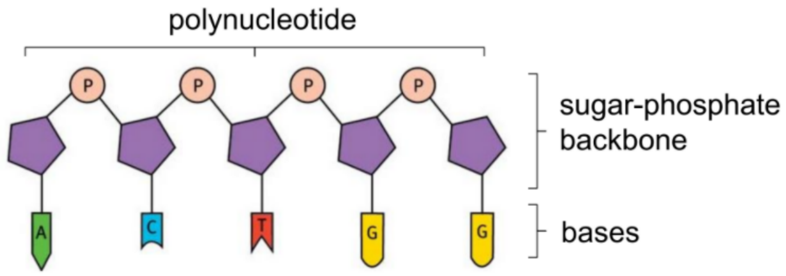
<u>Basic units of DNA</u>
Basic unit of DNA: **nucleotide**
1. **deoxyribose sugar**
2. **phosphate group**
3. **nitrogenous base**
    1) adenine (A)
    2) cytosine (C)
    3) guanine (G)
    4) thymine (T)



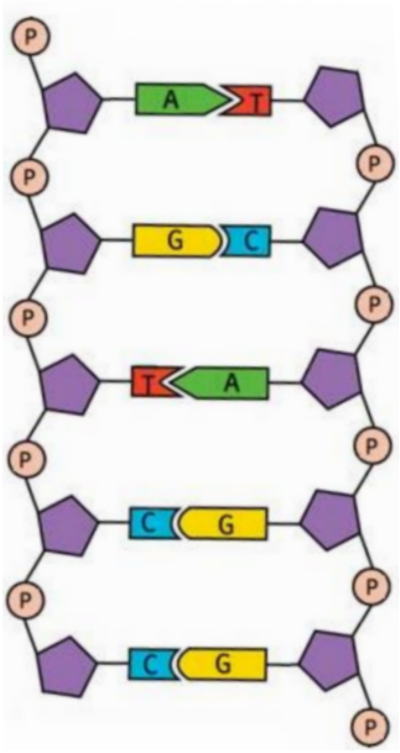
4 different nucleotide molecules

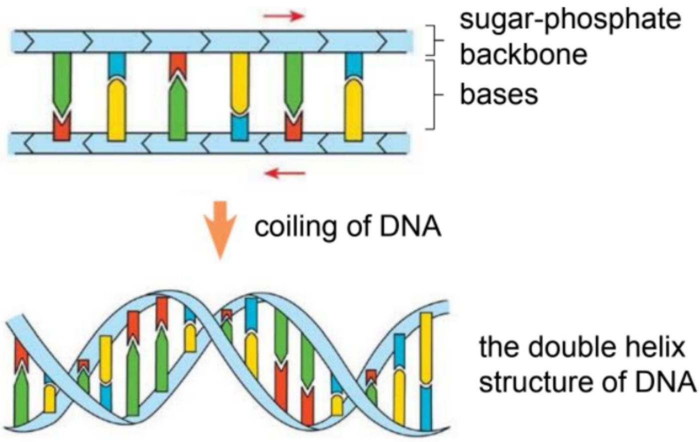


Polynucleotide: long chain of nucleotides



2 anti-parallel polynucleotide chains (run in opposite directions) → DNA molecule





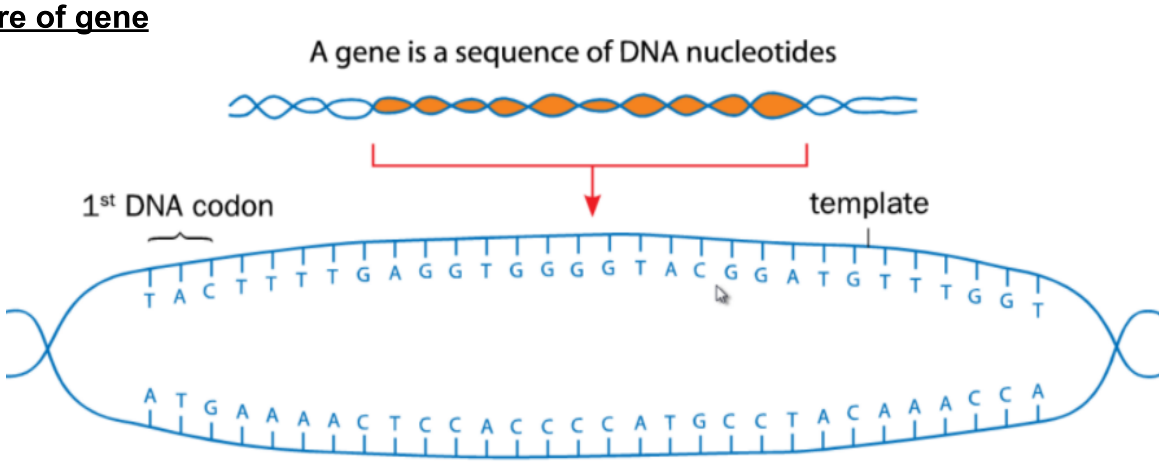
<u>**Rule of base pairing**</u>

Complementary bases: joined together by **hydrogen bonds**

1. A → T
2. C → G

20.2 Genes

<u>**Structure of gene**</u>

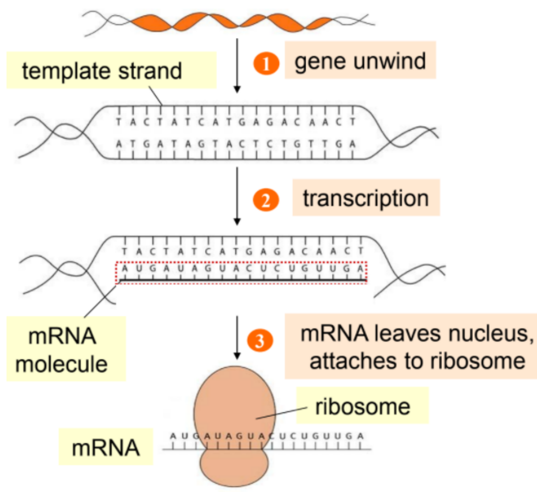


1 gene: segments of 2 polynucleotide chains (unzipped)
Template strand: 1 of the chains → determine type of protein made
Codon: 3 bases (code for 1 amino acid)

Note:
- 1 codon → 1 amino acid
- 1 gene → 1 polypeptide

Synthesis of polypeptides
Steps:
1. **Transcription**: DNA template → mRNA
2. **Translation**: mRNA codon → amino acids → polypeptide

Production of polypeptide

Step	Explanation	Figure
1. **Transcription**	• Region of DNA carrying gene unwind + unzip → separate 2 chains • mRNA made from template	
2. **mRNA**	• Nucleus → nuclear pore → ribosome (attach to)	
3. **Translation**	• Ribosome translate message in mRNA → amino acids (join together by peptide bonds → polypeptide) • Ribosome move along mRNA • End of mRNA strand: ribosome detach from mRNA → polypeptide released	

RNA: ribonucleic acid

Aspect	**DNA**	**RNA**
Sugar unit	deoxyribose	ribose
Nitrogenous bases	• adenine (A) • **thymine (T)** • cytosine (C) • guanine (G)	• adenine (A) • **uracil (U)** • cytosine (C) • guanine (G)
Structure of molecule	double-stranded	single-stranded
Ratio b/w bases	A:T = 1:1 C:G = 1:1	No fixed ratio
Existence	Permanent In nucleus	Temporary – made when needed Move out of nucleus

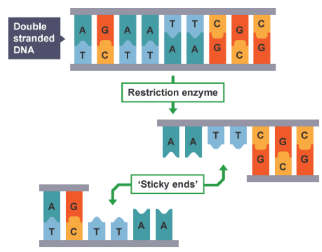
20.3 Genetic engineering

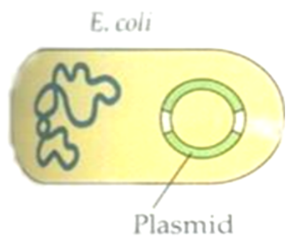
<u>Transfer human insulin gene → bacteria</u>

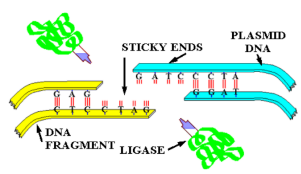
Insulin → treat type 1 diabetes

Pancreas of slaughtered animal	Genetic engineering
1. Diabetes develop antibodies against animal insulin after prolonged treatment → allergic to animal insulin 2. Diseases transmitted from animals → hmans	1. No allergic response → identical to human insulin 2. Easier + cheaper to produce in large quantities 3. Less risk of contamination by disease-causing microorganisms 4. No ethical concerns of vegetarians & religious groups

Materials required

Material	Explanation	Figure
1. **Restriction enzyme**	• Specific in action: recognise specific sequence of bases on DNA → restriction sites • Cut DNA only at these points → sticky ends	

2. **Vector**	• Another DNA molecule / bacterium / virus
	• Carry genes to be transferred → host cell
	• Come from plasmids:
	  ○ small, circular DNA molecules
	  ○ occur naturally in bacterial cytoplasm
	• Contain genetic markers: allow selection & identification of cells that have taken up vector-recombinant DNA molecule
		

3. **DNA ligase**	• Seal cut ends present in recombinant DNA molecules	

1 Isolate the desired gene

- Cut the gene using restriction enzymes.

↓

2 Insert the gene into the vector DNA

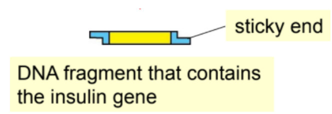
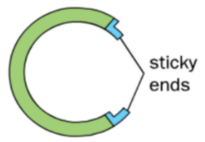
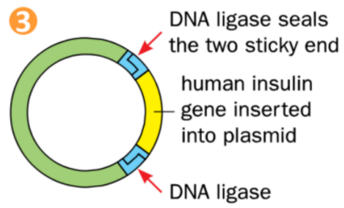
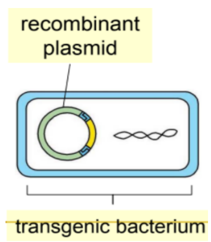
- Restriction enzymes that were used to cut the desired gene are used to cut the vector DNA.
- Both the cut vector DNA and gene are mixed together with DNA ligase, an enzyme that will help join the two molecules together.

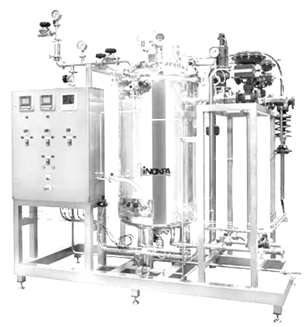
↓

3 Insert the recombinant plasmids into bacteria

- Mix recombinant plasmids with bacteria and heat- or electric-shock the cells.

Steps

Step	Explanation	Figure
1. **Sticky ends of human insulin gene**	• Obtain DNA fragment in human chromosome containing <u>insulin gene</u> • Cut gene using <u>restriction enzyme</u> → <u>sticky ends</u>	
2. **Sticky ends of plasmid**	• Obtain plasmid from bacterium • Cut plasmid with **SAME** restriction enzyme → sticky ends with bases complementary to that of insulin gene	
3. **Recombinant plasmid**	• <u>Mix</u> DNA fragment containing human insulin gene + plasmid → bind to plasmid (<u>complementary base pairing</u> b/w sticky ends) • Add <u>DNA ligase</u> → seal human insulin gene to plasmid → recombinant plasmid	
4. **Enter bacterium**	• Mix recombinant plasmid + <u>E.coli bacterium</u> • Apply <u>temporary heat / electric shock</u> → open up pores in CSM of bacterium → plasmid enter	

5. **Transgenic bacterium**	• Transgenic bacterium use new gene to make insulin → <u>isolated + grown</u> for mass production of human insulin
	• Supply optimum conditions for growth
	  ○ pH
	  ○ temperature
	  ○ nutrients
	  ○ oxygen
	• Insulin protein <u>extracted + purified</u> → used
		

<u>Transfer pest-resistant gene from bacterium to crop plant</u>

Steps

1. Use restriction enzyme → cut out gene from the bacterial DNA to produce sticky ends
2. Use same restriction enzyme → cut plasmid to produce complementary sticky ends
3. Insert the gene into plasmid
4. Insert recombinant plasmid into bacterium
5. Allow bacterium to <u>infect plant cells</u>
6. <u>Induce plant cells</u> to produce recombinant plants. A plant that has acquired a foreign gene is a **transgenic plant**.

Advantages & disadvantages

Advantage	Disadvantage
1. Increase food production 2. Use less pesticides → reduce environmental pollution	1. Insect pests develop resistance to poison produced by plant 2. Pest-resistance spread to weeds through cross-pollination 3. Kill useful insects 4. Produce herbicide-resistant crops → more effective destruction of weeds + kill insects → break links in food web & upset ecological balance

<u>Selective breeding & genetic engineering</u>

Comparison

	Selective breeding	**Genetic engineering**
Plants & animals used	closely related belong to same species	non-related species different species
Gene passed on to offspring	Defective genes	Genes selected → reduce risks of genetic defects passed on
Speed	Slow (breeding over several generations)	Fast (individual cells reproduce rapidly)
Space used	large amounts of land	small container
Efficiency	Less efficient (organism grow slower, require more food)	More efficient (transgenic salmon grow faster, require less food than ordinary)

<u>Benefits and issues of genetic engineering</u>

Benefits

Application	Benefit
1. Low-cost production of medicines	● Reduce cost → more affordable → more patients get access & treated
2. Produce crops that grow in extreme conditions ● drought-resistant crops ● salt-tolerant crops ● crops that make more efficient use of nutrients	● Grow crops – environmental conditions not suitable
3. Development of ● crops that produce toxins that kill insect pests ● pesticide-resistant crops	● X use costly pesticides – damage environment ● Bt gene from bacterium inserted into plants → produce toxin to kill insect pests

4. Development of foods designed to meet specific nutritional goals	● Improve nutritional quality (e.g. Golden Rice)

Social issues

Issue	Explanation
1. **New proteins in GM food**	● allergies
2. **GM food toxic / cancer-causing**	● modify gene → alter metabolic processes within plant → produce toxins ● serious health problems
3. **Loss of biodiversity**	● death of useful insects that feed on nectar of GM crop plants (e.g. honey bees, butterflies)
4. **Poor farmers**	● biotechnology company: engineer crop plants → produce seeds that cannot germinate ● farmers buy special seeds → financial burden
5. **Genes that code for antibiotic resistance**	● accidentally incorporate into disease-causing bacteria ● antibiotics ineffective in treating diseases
6. **Class divisions**	● only wealthy individuals can afford gene technologies

Ethical issue

Issue	Explanation
1. Vegetarians	● object consume + transfer animal genes → crop plants
2. Morally wrong	● exploit animals for medical research (designed to suffer) ● oncomouse: transgenic mouse introduced with gene that causes cancer → develop cancer for use in testing potential cancer drugs
3. Chemical / biological warfare	● deliberately create new combinations of genes

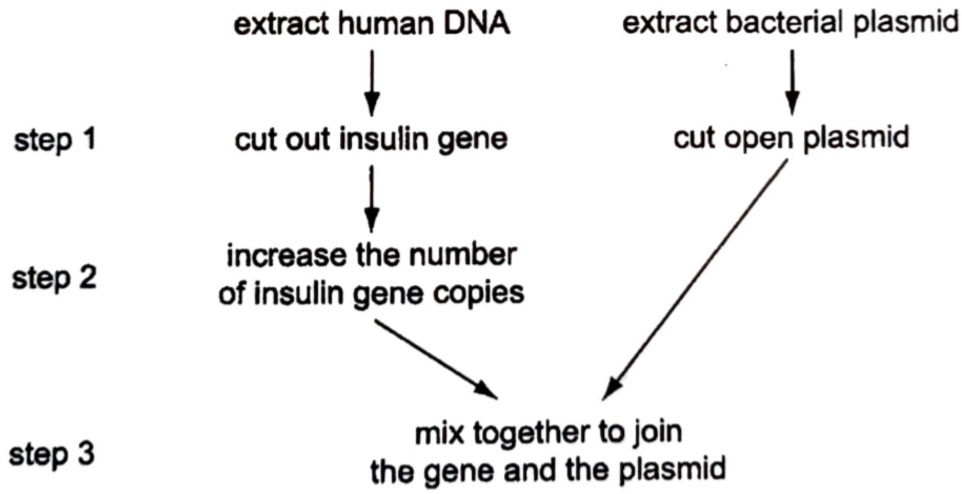
Typical questions

Multiple choice questions

1 Which statement applies **only** to a gene? (N2011/P1/Q34)

A It can be copied during cell division.
B It can control multiple characteristics.
C It codes for a single polypeptide.
D It is composed of four bases.

2 The diagrem outlines part of the process to produce recombinant DNA that will synthesise human insulin.
At steps 1, 2 and 3, enzymes are involved.



Which row correctly identifies the enzyme in each step? (N2011/P1/Q35)

	step 1	step 2	step 3
A	polymerase	ligase	restriction
B	polymerase	restriction	ligase
C	restriction	ligase	polymerase
D	restriction	polymerase	ligase

3 Which statement about DNA is correct? (N2012/P1/Q35)

A A molecule of DNA contains many genes.
B A molecule of DNA is larger than a chromosome.
C A molecule of DNA is the same size as a gene.
D Each molecule of DNA is always the same length.

4 Which statement describes a gene? (N2013/P1/Q35)

- **A** a number of DNA molecules
- **B** a pair of alleles
- **C** a sequence of nucleotides
- **D** the chain of alleles on a chromosome

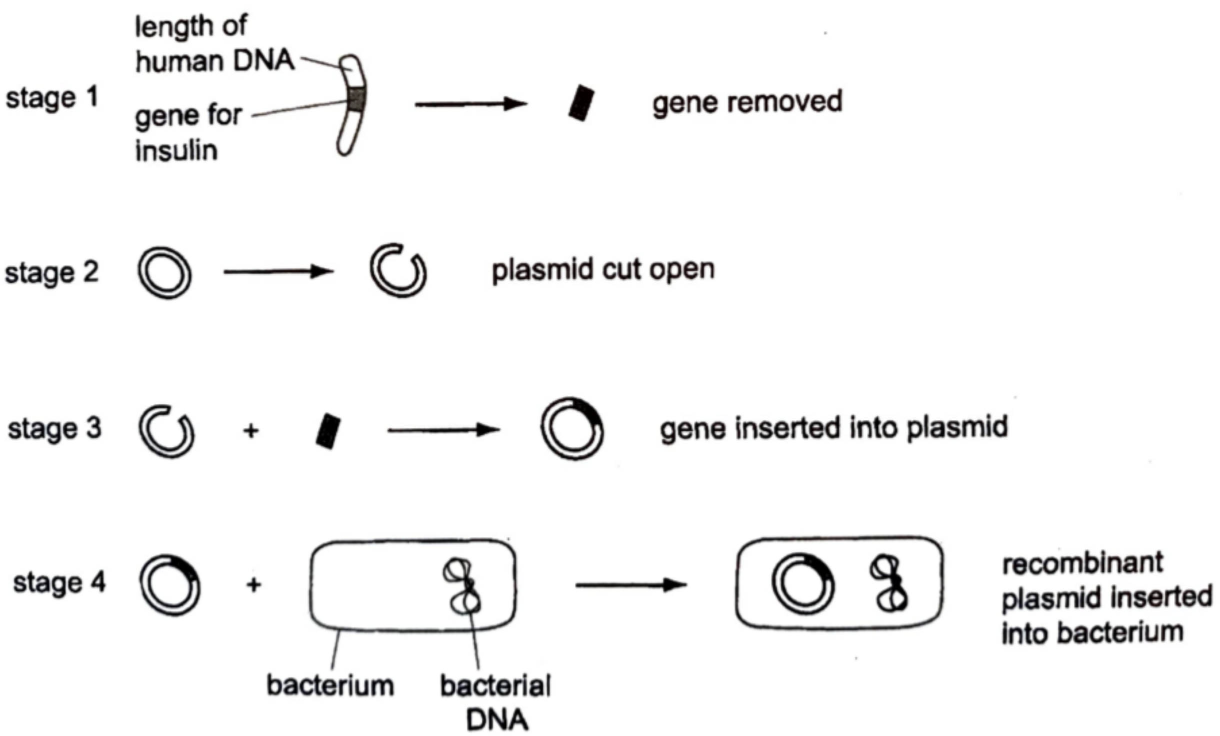
5 Which statement describes a gene? (N2017/P1/Q34)

- **A** a base with a sugar and phosphate group
- **B** a number of DNA molecules
- **C** a sequence of nucleotides
- **D** the chain of alleles on a chromosome

6 What defines a gene? (N2019/P1/Q34)

- **A** the sequence of amino acids found in the DNA that codes for a polypeptide
- **B** the sequence of amino acids found in a polypeptide
- **C** the sequence of nucleotides found in a polypeptide
- **D** the sequence of nucleotides found in the DNA that codes for a polypeptide

7 A human insulin gene can be cut out of human DNA and inserted into the plasmid of a bacterium. The diagrams show four stages of this process.



Which row correctly identifies the enzyme used at one of these stages? (N2014/P1/Q35)

	stage	ligase enzyme	restriction enzyme
A	1	used	not used
B	2	not used	used
C	3	not used	used
D	4	used	not used

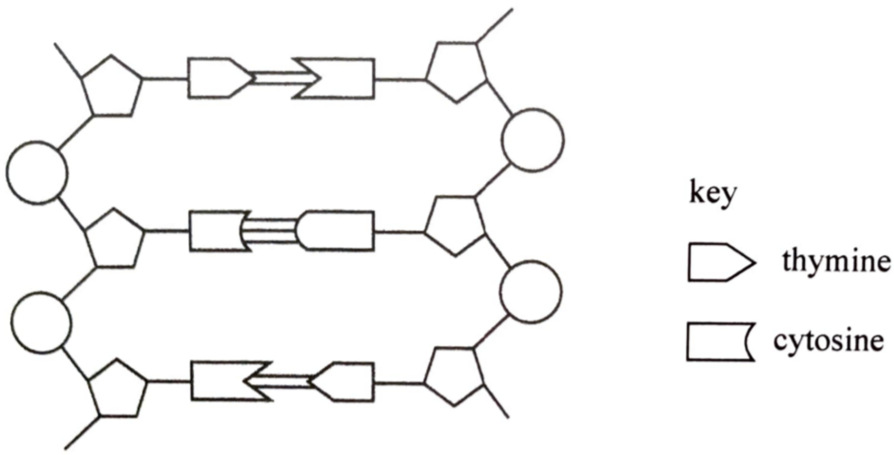
8 What describes the genes in a transgenic organism? (N2015/P1/Q35)

A derived from two organisms
B have mutated naturally
C inherited from two individuals
D modified by radiation

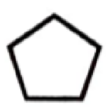
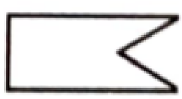
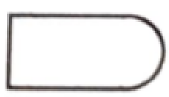
9 Which statement describes a transgenic plant? (N2016/P1/Q35)

A a plant containing artificially inserted genes from a plant of a different species
B a plant in which some of the DNA has been removed
C a plant which has acquired a new gene by mutation
D a plant which has been bred by selection and controlled breeding

10 The diagram below shows a short section of a molecule of DNA.



Which row identifies the other shapes used in this diagram? (N2016/P1/Q34)

	shape			
				

A	phosphate	sugar	adenine	guanine
B	phosphate	sugar	guanine	adenine
C	sugar	phosphate	adenine	guanine
D	sugar	phosphate	guanine	adenine

11 A transgenic maize plant produces a chemical that kills the insects feeding on it.
Which implications of this situation are correct? (N2017/P1/Q35)

	insects beneficial to other plants may be killed	food chains can be disrupted	less weed killer is used by farmers
A	no	yes	yes
B	yes	no	yes
C	yes	yes	no
D	yes	yes	yes

12 Scientists used genetic engineering to introduce a gene for producing fluorescence into an animal. Some of the steps in the process are listed.

1. DNA is used to produce proteins coded for by the inserted gene.
2. Restriction enzymes are used to cut out the gene.
3. The gene coding for fluorescence is identified.
4. The gene is inserted into host DNA.

What is the correct order for these steps? (N2018/P1/Q34)

A $1 \rightarrow 3 \rightarrow 2 \rightarrow 4$
B $2 \rightarrow 3 \rightarrow 4 \rightarrow 1$
C $3 \rightarrow 2 \rightarrow 4 \rightarrow 1$
D $4 \rightarrow 2 \rightarrow 1 \rightarrow 3$

13 Which process would result in a transgenic organism? (N2019/P1/Q35)

A adding a chemical to embryo plants that causes the diploid chromosome number to double which makes the plant produce larger fruit
B crossing two different varieties of the same species of plant to obtain high yield crops resistant to insect pests
C fusing an egg cell without a nucleus from one animal with a diploid cell from a related species
D inserting a gene from one species into the egg cell of a different species to make the animal produced grow faster

14 Which mode of information transfer usually does not occur?

A DNA to DNA
B DNA to RNA
C DNA to protein
D all occur in a working cell

Structured questions

1 Define the term *gene*. [2]
(N2012/P2/A1a)

A gene is a small segment of DNA, located at a particular locus on a chromosome, which stores hereditary information and has a specific function. Each gene contains the information to make a protein.

2 Medical biotechnology can be used to produce chicken eggs that contain human proteins. These proteins can be used in medicines.
Discuss **two** social and **two** ethical implications of using chickens to produce human proteins. [4]
(N2019/P2/A5)

Social implications:

- Vegetarians may object to consuming medicines made by animals as it could be viewed as similar to consuming meat.
- There are also concerns that this could lead to class distinctions as only wealthier individuals could afford such medicines which are likely to be sold at a higher price.

Ethical implications:

- Some feel that it is morally wrong and against animal rights in using chickens to produce human proteins as it often involved the killing of these chickens during the extraction process.
- The introduction of a foreign gene in the chicken may also lead to health issues for the transgenic chickens, violating animal rights and causing unnecessary suffering.

3 (N2013/P2/B9a, b)

(a) Describe what is meant by a *transgenic organism*. [3]

- A transgenic organism is any organism that acquires a foreign gene from other organisms of the same / different species.
- These genes are artificially inserted into the organism.
- For example, human insulin gene is artificially inserted into Escherichia coli to form transgenic bacteria to produce insulin.

(b) Suggest three reasons for the development of transgenic organisms. [3]

- Genetically engineering pest-resistant crops + increasing crop yield + increase food production.
- Genetically engineering organisms to contain more nutrients + benefit people that feed on them + for example genetically engineered production of Golden rice that is infused with β-carotene (used to make vitamin A).
- Transgenic organisms with favourable traits such as increased milk production in cows to be sold in the market + increased profit for farmers.
- **Mass** production of useful chemicals such as insulin + lower cost of medicine production + making medicine cheaper for patients.

4 (N2017/P2/B10 EITHER)

(a) Describe the structure of DNA. [6]

- DNA found in the nucleus of cells contains genetic information that determines the characteristics of a person.
- Each DNA strand is made from basic units known as nucleotides. Each nucleotide consists of three parts: a deoxyribose sugar, a phosphate group and a nitrogenous base. There are four types of nitrogenous bases – adenine, thymine, guanine and cytosine.
- The nucleotides join together to form a long chain of polynucleotides (a DNA strand).
- Two strands of DNA are joined together by complementary base pairing of specific nucleotides to form the double-helical structure of DNA. Adenine always forms hydrogen bonding with thymine; guanine always forms hydrogen bonding with cytosine. The sugar-phosphate backbone of the DNA strands face outwards, while the nitrogenous bases face inwards. The two strands of DNA run in opposite directions.
- The DNA molecule is coiled around proteins and further supercoiled to form chromosomes.

(b) Explain how a transgenic organism can be produced. [4]

- A transgenic organism is an organism which has acquired a foreign gene. For example, human insulin gene can be transferred from human cells to bacterial cells in order for the bacteria to produce insulin. The bacteria are known as transgenic organisms.
- The human insulin gene is first identified and cut out using an appropriation restriction enzyme. The same restriction enzyme is used to cut a plasmid, a circular piece of DNA found in bacteria, to produce DNA with complementary sticky ends.
- The insulin gene is then inserted into the plasmid using DNA ligase. The bacteria are treated with electric shock to take up the plasmid containing the insulin gene.
- The bacteria which have taken up the plasmid will multiply and be identified. These bacteria are then isolated and grown in fermenters under optimal conditions.

5 A gene in a piece of DNA contains 6000 nucleotides (or bases). It codes for the production of a protein.

Deduce the number of amino acids that would be required to produce the protein. Show your working below. [2]

No. of nucleotides on template strand (1 polynucleotide chain) = 6000 / 2 = 3000

No. of DNA codons on template strand = 3000 / 3 = 1000

No. of amino acids required = No. of DNA codons = **1000**

6 (N2009/P2/Q10 EITHER)

(a) Outline the relationship between DNA, genes and chromosomes. [3]

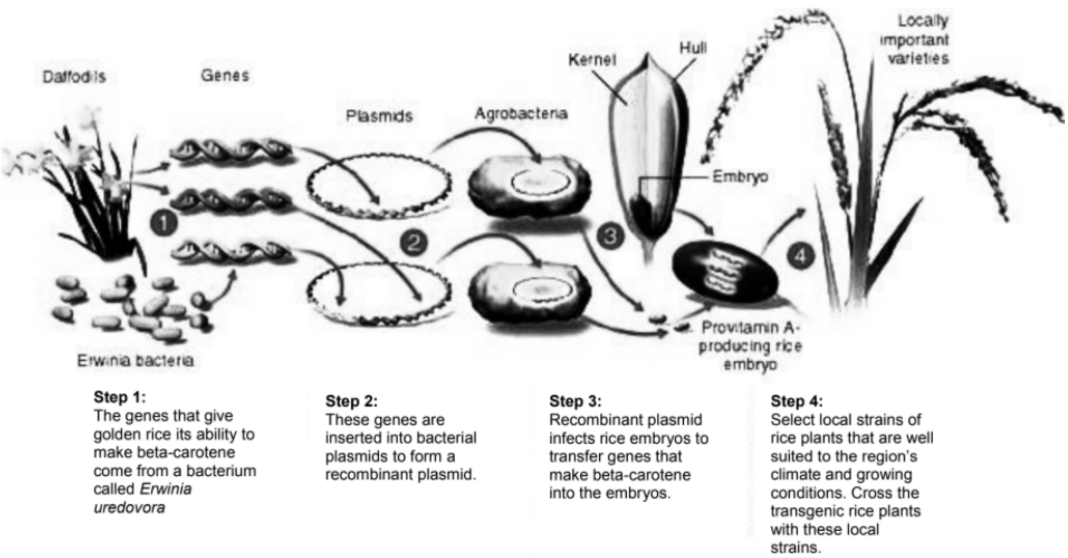
1. **DNA** winds around proteins to form chromosomes.
2. Each **chromosome** is made up of DNA and it carries many genes along its length.
3. **Gene** is a unit of inheritance borne on a particular locus of a chromosome + made of a sequence of DNA nucleotides that code for a polypeptide to make a protein.

(b) State the significance of the order of bases in a DNA molecule. [2]

- The order of DNA nucleotide bases determine the type of amino acid
- and the production of a specific polypeptide/protein

7 Rice is the staple diet in many parts of the world. It lacks a number of important nutrients, including β carotene, from which vitamin A is synthesised. Adequate concentrations of vitamin A give protection against night blindness.

Golden rice, which contains high levels of β carotene, was developed by genetically modifying rice using genes from a daffodil (a flowering plant) and bacterium. The figure below shows the steps taken.



(a) Briefly describe how steps 1 and 2 in the figure above can be carried out. [5]

1. Use a restriction enzyme to cut genes that code for β carotene from Erwinia bacteria & the daffodil
2. Using the same restriction enzyme, cut the bacterial plasmid
3. to produce sticky ends with complementary bases
4. Mix cut genes with cut plasmid
5. seal genes to plasmid using DNA ligase to form a recombinant plasmid

(b) An agreement was made between the commercial company that owns the production rights of golden rice and its developers. This allows the developers to give the rice to government-run breeding centres in rice-dependent countries. Local farmers will be able to grow the rice without paying a high fee.
Suggest why governments in rice-dependent countries are in favour of golden rice. [1]

- Local farmers can sell their rice for a profit without paying a high price for it, promote the economy of country.
- Increasing the nutrient content of rice by infusing it with β-carotene can reduce the risk of night blindness among people.
- Reduce malnutrition in developing countries.

(c) Discuss the ethical implications of genetic engineering. [4]

1. Exploitation of animals for genetic engineering
    - exploited for food and development of medical products (e.g. unregulated production of growth hormones causes harmful effects on the health of animals, making them more susceptible to diseases
    - concerns on whether animals are biologically capable of withstanding the additional stress of increased production of milk, meat and other products.
    - experiments that causes animals to suffer (e.g. oncomouse in cancer drug research develops tumour, suffers and dies)

2. Cloning of mankind
    - When Man is given control over life, being able to choose desirable traits in offspring → this might lead to class distinction in human