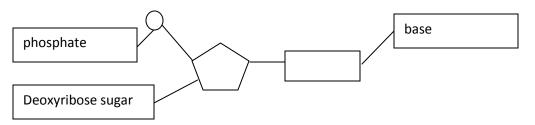
Section 1 – The structure and replication of DNA

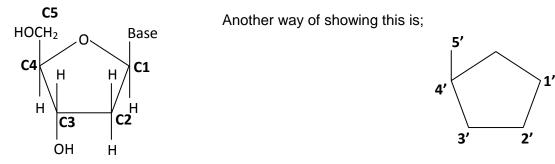
Structure of DNA

DNA = deoxyribonucleic acid

The basic subunit of any nucleic acid is the nucleotide



A closer look at the sugar allows you to number the carbons.



A double strand of DNA is produced by;

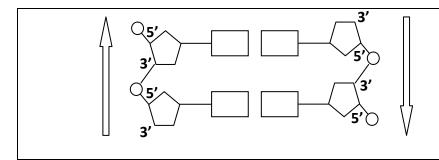
Strong covalent bonds between the phosphate of one nucleotide and the sugar of the next (the backbone)

And weak hydrogen bonds between complementary base pairs.

There are four bases ; adenine (A), thymine (T), guanine (G) and cytosine (C)

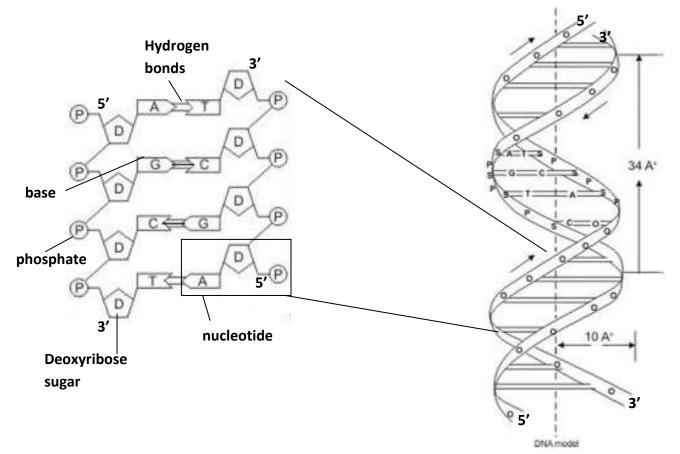
The base pairing rule is that A pairs with T, G pairs with C

Draw two nucleotides, join the 3' carbon of the top nucleotide onto the 5' end of the next (the phosphate). Use the simplest form of the nucleotide (top of the page)



Now add a complementary strand. Tricky bit, it runs 3' to 5' (the pentagons will be upside down!)

Label these diagrams with; sugar phosphate back bone, 5' and 3' ends of each strand, hydrogen bonds, nucleotide, deoxyribose sugar, phosphate, base. (and anything else you think is useful)



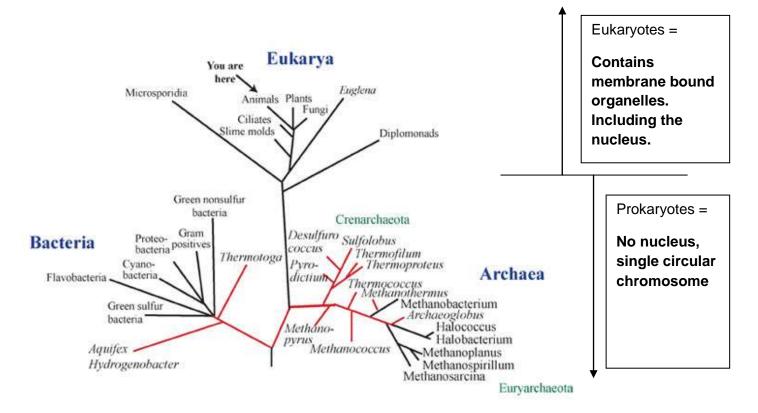
DNA is a double stranded, double helix with antiparallel strands.

Experimental Evidence for the Structure of DNA.

- 1. Griffith worked with bacteria and mice. Showed that there was a way of passing on lethality in different strains of bacteria called the process **transformation**
- Avery et al continued work on the transforming principle. They stated that it was DNA fragments that were needed for transformation. (some scientists were not convinced and continued to back protein as the molecule for inheritance)
- 3. Hershey & Chase worked with bacteriophage (viruses that attack bacteria). They used radioisotopes to track proteins and DNA in the virus. Showing categorically that it was DNA that was coding for the viral information.
- Chargaff Chargaff worked on studying the ratios of bases is different organisms. He found that A=T and G=C, although the proportions of each varied from organism to organism
- 5. Franklin and Wilkins published detailed x-ray diffraction data on crystallised DNA.
- 6. Watson and Crick published a structure of DNA which fitted all the data.

Organisation of DNA

Phylogenetic tree of all life on Earth



Prokaryotes contain circular chromosomes and can also contain smaller circles of DNA called plasmids (you have heard about these in genetic engineering of bacteria to make insulin).

Eukaryotes contain linear chromosomes, strings of DNA wrapped around proteins called histones, all contained in the nucleus. Some organelles; Chloroplasts and Mitochondria, also contain DNA, these are small circular plasmids.

Extra; How does this fact back up the endosymbiotic theory of cell ultrastructure?

Cell characteristics	Prokaryote	Eukaryote
Contains nucleus	No	Yes
Chromosomal DNA	Circular	Linear
Plasmids in cytoplasm	Yes	No
Contains organelles with circular DNA	Νο	Yes (chloroplast, mitochondria)

Replication of DNA

Semi-conservative replication means half of the new DNA comes from the old DNA (you have kept 1/2 of

the original DNA).....

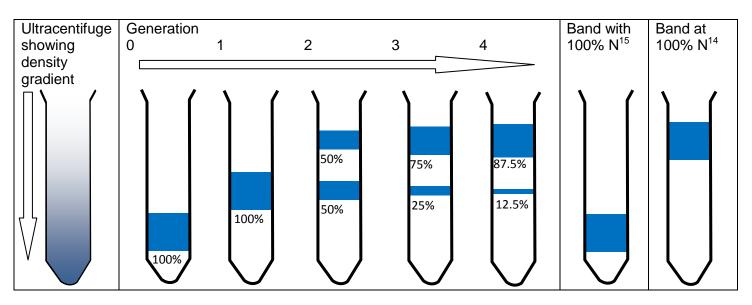


Diagram of results from Meselson-Stahl experiment using $N^{\rm 15}$ and $N^{\rm 14}$ labelled bases

Explain how these results confirm semi-conservative replication

The first band contains DNA all of the same mass. N15 DNA.

Incubated with only access to N¹⁴

In the second generation it all masses the same, but at mid way between N^{15} and N^{14}

In the third generation half of the DNA gets N^{14} , so you get all N^{14} , the other half still has half N^{15} /half N^{14}

By the third you are now getting more changed to fully N¹⁴

...

Extra; find out how Herbert Taylors experiments in 1958 replicated Meselson-Stahl's conclusions

Extra; the other postulated mechanisms were; conservative replication and dispersive replication – find out what each means.

Reason you need it
Provides the code to copy.
To produce the new strand of DNA
To provide the enzymes with energy to; unwind the DNA, pair up the new nucleotides, join up the backbone, rewind the DNA spiral
Unwind the DNA (and hold replication fork open)
Pair up the free nucleotides
Join up the Okazaki fragments
To allow attachment of DNA polymerase

Complete the following page with descriptions of each stage of replication. Include labels or notes on all of the requirements and also; Okazaki fragments, DNA polymerase direction, leading strand, lagging strand

What is meant by 'directionality of DNA polymerase' **DNA polymerase can only add nucleotides running** in one direction.

Leading Strand = the DNA strand that is added to continuously (attaching to the 5'end)

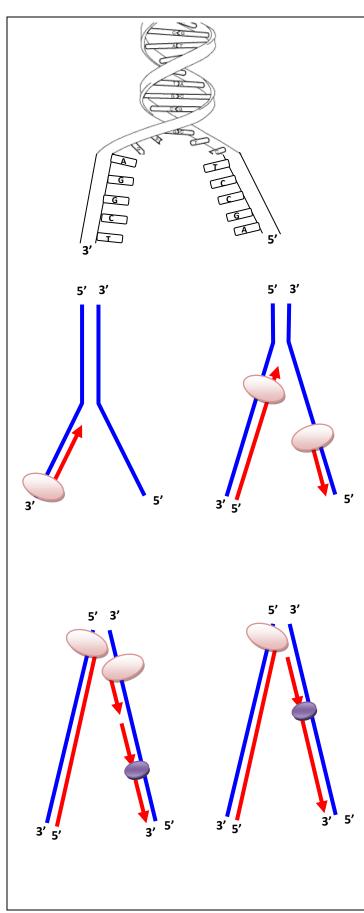
Lagging Strand = the DNA strand that can only be added to once the spiral opens up far enough for the enzyme to lock on

Okazaki fragments = the DNA fragments that form on the lagging strand

Replication fork = the point where the DNA spiral is 'unzipped'

On long chromosomes several replication forks might be started at various points along the length of the chromosome. Why would this be a good idea?

Cuts down the length of time it will take to complete replication of the whole length.



DNA untwists and 'unzips. Requires enzymes (helicase) and energy (ATP).

Gives you two ends and a replication fork.

DNA polymerase can only read $3' \rightarrow 5'$, so attaches to the 3' end creating the <u>leading</u> strand.

This will replicate continuously.

Adding complementary nucleotides to the chain.

As the replication fork moves DNA polymerase can attach and read back down to the 5' on the other strand (<u>lagging</u> <u>strand</u>)

The DNA polymerase drops off the end

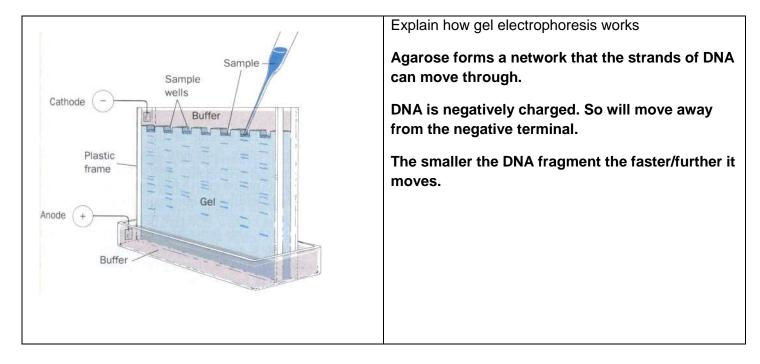
As more of the lagging strand becomes available more fragments are formed. (Okazaki fragments)

These fragments are joined together by the enzyme <u>ligase</u> creating a continuous strand.

At the end you have two identical strands, each containing one strand from the original molecule =

Semiconservative replication

Gel Electrophoresis - separating DNA



Polymerase Chain Reaction (PCR) – Using DNA replication to amplify a specific section of DNA

Stage	Temperature (°C)	What is happening
1	92	DNA strands are denatured (pull apart)
2	55	Primers anneal
3	72	Taq polymerase activity

In Stage 1 why do the strands separate instead of just disintegrating? Hydrogen bonds are weaker than covalent bonds, so the base pairing will break before the backbone.....

In Stage 2 what is the function of the primers? **To bracket the sequence and allow DNA polymerase to** attach.

In Stage 3 Why is *Taq* polymerase used? **Originally extracted from a thermophilic bacteria – will not be denatured by the high temp.**

Looking for a specific sequence of DNA on a chromosome could be called a 'needle in a haystack' search. It has been stated that the primers are a needle from which a haystack can be grown. Explain these statements.

Extra; find out how PCR is used in DNA fingerprinting

Section 2 – Gene Expression

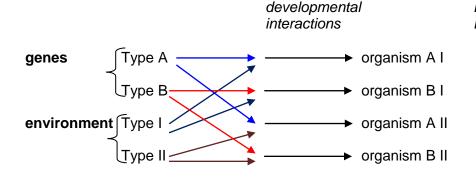
Genes are DNA sequences that code for particular proteins. Although every individual will have genes for the same proteins, there may be different forms of the gene, these are called <u>alleles</u>. In humans, since you have two copies of each chromosome you will have two copies of each gene. The copies may or may not be the same form.

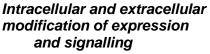
Genotype means the alleles present

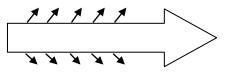
Phenotype means the appearance of an organism

Determination of Phenotype

A model showing how genes and environment interact





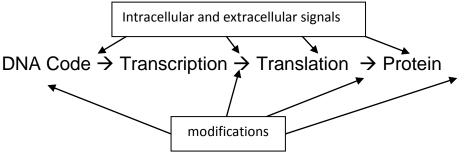


Explain what is meant by Epigenetic modification

Changes on the DNA that do not change its sequence but may affect the expression of the DNA. E.g. methylation of bases may cause a particular gene not to be expressed so changing the phenotype from dominant to recessive.

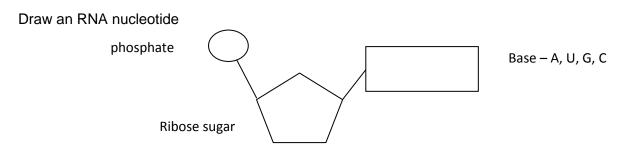
Control of gene expression

The human genome consists of more than 3 billion base pairs. The DNA can be split into coding and noncoding DNA. The coding DNA (less than 2%) produces proteins, the non-coding DNA is involved in the gene expression.



Structure and function of RNA

RNA ribonucleic acid



Differences between DNA and RNA

	DNA	RNA
Found in	Nucleus	Nucleus/ cytoplasm
Strands	2	1
Sugar in nucleotide	Deoxyribose	Ribose
Adenine paired to	Thymine	Uracil

Forms and functions

	Stands for	Found in	Function
mRNA	Messenger RNA	Nucleus & Rough ER / cytoplasm	Carry copy of genetic code from DNA to ribosome
tRNA	Transport/ transfer RNA	Cytoplasm/ ribosome	Carry amino acids from cytoplasm to ribosome
rRNA	Ribosomal RNA	Ribosome	Take part in translation of mRNA to amino acid code

<u>Codons</u>

Nucleotide code only has 4 letters. Protein 'code' has 20.

Explain what is meant by this statement

There are only 4 letters in any nucleic acid (ATGC or AUGC), but there are around 20 different amino acids

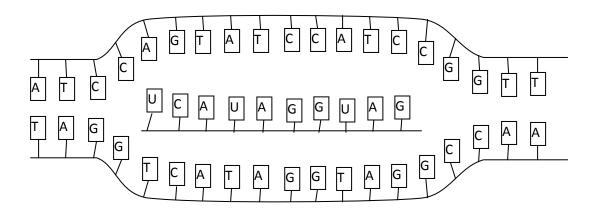
Explain why you need a codon length of 3 nucleotides for each amino acid code.

If you only had a code length of 1, there are only 4 combinations (4^1) , code length of 2 gives you 16 (4^2) , code length of 3 gives you 64 (4^3) – more than the 20 you need, but 16 is not enough.

Transcription

- Section of DNA (gene) uncoils
- RNA nucleotides pair up with DNA nucleotides
- Strand of **mRNA** released

Complete the diagram below adding in a mRNA molecule and completing complementary base-pairing on the DNA and RNA

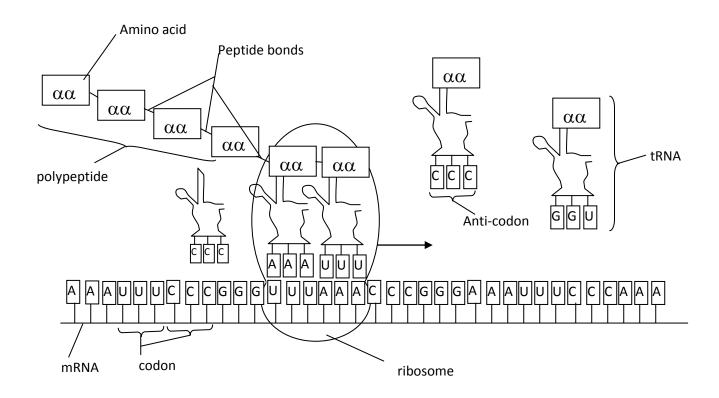


This produces a section of mRNA called the **primary** transcript.

The primary transcript is converted to a **mature** transcript by a process called RNA **splicing**.

This removes sections called **introns** and joins together the coding sections called **exons**.

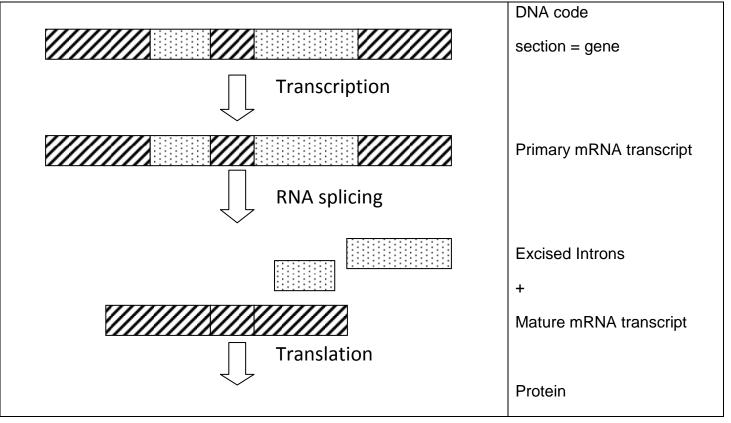
Translation



Describe what is happening in the diagram above

Ribosome covers 2 codons of mRNA at a time- tRNA carrying the amino acid specific to its anticodon pairs up with codons. Peptide bond joins amino acids on top of tRNAs – the first tRNA is released and the ribosome moves along a codon – process repeated

Overview

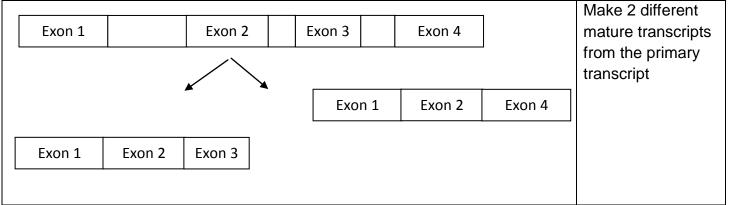


One gene – many proteins

The number of genes coded for in DNA is less than the number of different proteins you find in organisms.

There are 2 main mechanisms for giving multiple proteins from a single gene.

1. Alternative RNA splicing



2. Post-translational modification (explain and give an example)

Altering the protein structure after you have produced the amino acid chain.

e.g. removal of a section of polypeptide changes trypsin from inactive to active form.

e.g. adding on phosphate groups can change proteins from inactive to active forms.

.....

Shapes of proteins

Primary structure (1°)

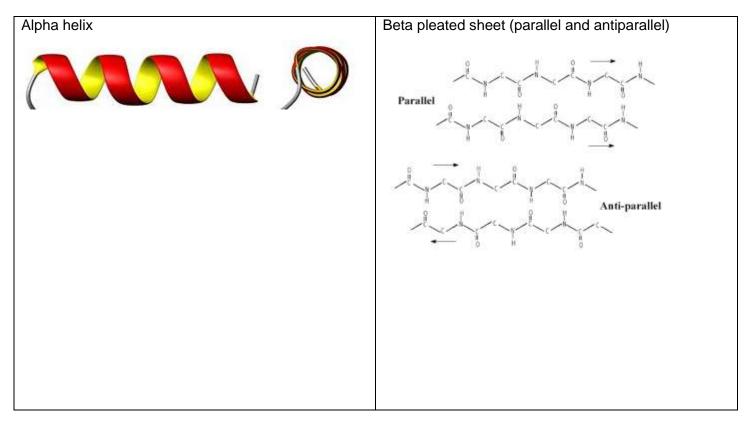
Consists of the initial amino acid sequence, linked by strong covalent bonds.

Crosslinks can be by **covalent** disulphide bridges and **ionic** interactions between charged amino acids.

There are also weaker hydrogen bonds and other London dispersion forces.

Secondary structure (2°)

There are two main secondary structures stabilised by **hydrogen** bonds.

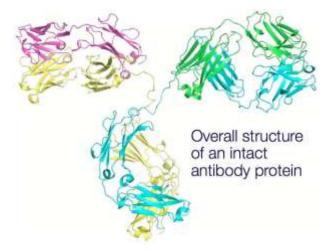


Tertiary structure (3°)

The final folded shape of the polypeptide is the tertiary structure.

Quaternary structure (4°)

Polypeptides are linked together (each chain forms a domain) and sometimes other non-protein elements are added.



Types of Protein

Туре	Basic Structure	Example	Properties
	Regular structure	Keratin	Strong inelastic
Fibrous		Collagen	Structural / support
	Folded	Any enzyme	Very specific shape
Globular		Antibodies	Very specific to particular antigens
Conjugated	Protein + non- protein component	haemoglobin	Association and dissociation of Oxygen under differing conditions
Conjugated		chlorophyll	Can absorb light energy to fix as chemical in photosynthesis

Cellular differentiation

Cells that carry out specific functions have specific shapes and biochemical pathways. That means they need to be altered by switching on and off genes at specific points in time and at particular points within an organism.

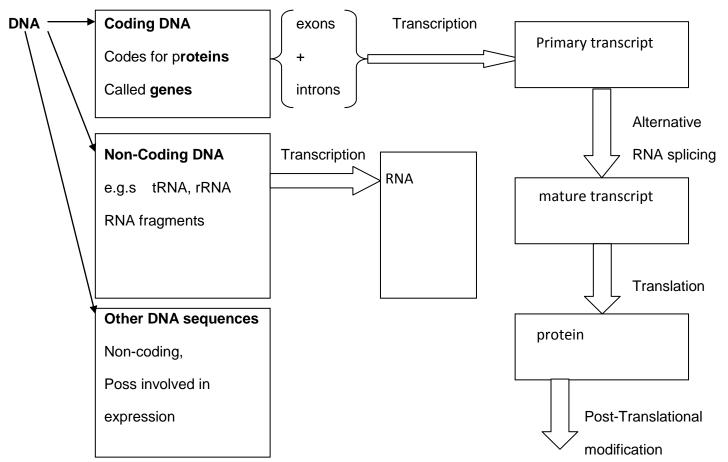
Using knowledge from the structures in these cells, write notes on what genes would need to be switched on and off (try and think about order as well)

Cell type	Red blood cell in human	Xylem tube in plant	Pallisade mesophyll cell in leaf
Function	Carry oxygen	Carry water	Photosynthesis
Genes to alter from an undifferentiated cell	-Express haemoglobin gene -create shape in cell (biconcave disk) - destroy nucleus	lignin deposition connect end walls destroy end walls	Production of chloroplasts Chlorophyll production Enzymes for photosynthesis Cell wall shape to give close packing with other cells

DNA & tl	he Genome	Higher Biology			
<u>Differen</u>	itiation in plants				
Occurs	Occurs at Meristems = regions of unspecialised cells that can undergo division				
Apical n	neristems are found at the tips of plants (root or shoot)				
and incr	rease length				
Lateral ı	meristems are found in the stems of plants				
and incr	rease girth				
<u>Differen</u>	itiation in animals				
Stem ce	ell cells that are capable of cell division and differentiation				
•	otent (definition) can develop into a sub-set of cell types e.g.				
Totipote	ency (definition) can develop into any cell type.				
Embryo	nic stem cells are totipotent Tissue (adult) stem cells are pleur				
<u>Stem ce</u>	ell research and implications				
Make su	ure that you have notes that cover;				
- 3	Sources of stem cells \Box				
- 3	Stem cell use for repair of damaged tissue / organs (therapeutic	use) 🗖			
- 3	Stem cell research into cell processes \Box				
- (Stem cell research into disease \Box				
- (Stem cell research into drug use and development \square				
- {	Ethical considerations in use of stem cells \Box				

Section 3 – The Genome

Structure of the Genome



Mutation

Mutation means an alteration in the DNA.....

.....

This shows some data on mutation in corn. Calculate the average number of mutations per million.

Gene	Number of gametes tested	Number of mutations	Average number of mutations per million gamete
R→r	554,786	273	492
I→i	265,391	28	106
Su→su	1,678,736	4	2
Sh→sh	2,469,285	3	1

Why is it necessary to convert to 'per million'? to give a fair comparison with the different numbers involved.

What can you say about mutation frequency...it is not very high, but different genes have quite different levels of mutation

Testing the mutation rate

To even see a mutation, you need it to have a measureable effect. These are some types of mutation. An example has been given for each one – work out what is meant by the term. (problem solving)

Morphological	e.g	Dwarf pea plants compared to normal pea plants.	A change in the shape
Biochemical	e.g	A class of fungi only grow if there is adenine in the culture medium	A change in the internal chemistry
Resistance	e.g	Some bacteria are no longer killed by common antibiotics	A change in how it reacts to a chemical that would normally kill it.
Hypermorph	e.g	Form of <i>Penicillium</i> that produces large quantities of the antibiotic penicillin.	Starts making much more of something it normally makes
Hypomorph	e.g	Form of Drosphilia that has light red eyes rather than bright red (wild- type)	Starts making a lot less of something it normally makes

Single Gene mutations

Reading frame = the triplets that the nucleic acid is split into (there are 3 possible frames)

Specific terms are used to describe; how a mutation has taken place and the effect it has.

Substitution – e.g. Original mRNA Amino acid Substitute G→A		Definition Base(s) are switched for another. In this case a G for an A
Amino acid	γγγ	
Deletion – e.g.		Definition
Original mRNA	çecennncc	Base is deleted, will shift the reading frame and all amino acids
Amino acid		'downstream'.
Delete of G	CGCUUUCCA	
Amino acid	᠋᠋᠆᠆᠆᠆᠆᠆᠆	
Insertion – e.g.		Definition
Original mRNA	cecennncc	Pass is added in will shift the reading frame and all amine aside
Amino acid	ᡃ᠆ᠰ᠆ᢇ᠆ᠰ᠆	Base is added in, will shift the reading frame and all amino acids 'downstream'.
Insert of A	ÇGÇ AG YUUÇC	
Amino acid	᠂᠆ᠰ᠆ᠰ᠆ᠰ᠆	

DNA & the Genome			Higher Biology	
Forms of mutation				
Point mutations means only a single point in the amino acid code is changed.				
	nutation means the reading	-	ed, so all code downstream of mutation is	
Describing so	ome point mutations;			
Missense				
mRNA		mutation		
amino acid	phe leu thr aspn		phe pro thr aspn	
Called <i>misse</i>	<i>nce</i> because the amino aci	d code has bee	en altered to a different amino acid	
Nonsense				
mRNA	CGUAGUUAUGGC	mutation		
amino acid	gly ser tyr gly		gly ser STOP	
	<i>ns</i> e because the change in		a stop codon. So making no sense amino acid	
Silent				
mRNA	CCUGAUGAAGGC	mutation	CC C GAUGAAGGC	
amino acid	pro asp glu gly		pro asp glu gly	
Called <i>silent</i> because the change in code creates a code that gives the same amino acid on translation.				
Splice-site – Explain how a mutation in a splice site could affect proteins.				
This could alter the protein by either stopping a splice or adding one in. Creating a totally different mature transcript from the primary.				
Repeats – Explain how nucleotide repeats can affect protein code.				
Increasing a repeat could cause a frame-shift or increase the number of a particular amino acid(s)				
inside a protein, so altering the structure				

Chromosomal mutations

During gamete formation (meiosis) the pairs of chromosomes line up on the equator. This is the point where you get mutations involving changes to the chromosome structure.

Duplication

Definition genes from one homologous chromosome are moved into the other, creating duplication of the gene on the chromosome. (the other homologous loses and may no longer function correctly – see deletion)

Explain how duplication can cause mosaic transmission of a condition like Downs Syndrome

Instead of getting a whole chromosome, only some of the genes are duplicated, this can still lead to 3 copies of a gene (2 from duplication and one from normal chromosome), creating some of the symptoms of Down's

Deletion

Means a gene or genes is deleted from the chromosome

Explain how this could change expression of recessive alleles if a dominant form of the gene is deleted, then what ever allele is on the homologous chromosome will show in phenotype i.e. recessive will only need one copy.

Translocation

Means section of chromosome is moved from one to another

Explain how translocation can cause mosaic transmission of a condition like Downs Syndrome **a section of** chromosome 21 moved onto another chromosome, gives 2 normal chromosome 21 + the additional section making some genes available in triplicate i.e some symptoms of Down's

Non-Disjunction.

Caused by failures in the spindle fibres during meiosis. A single failure causes trisomy of one chromosome.

In humans trisomy of chromosome **21** causes Downs Syndrome. Failure of a complete set of spindle fibres

will create a cell with multiple extra sets of chromosomes, this is called **polyploidy**

Key point; Mutations increase variation. Variation is the raw material for evolution.

DNA & the Genome

Mutation and evolution

Polyploidy

The number of chromosomes found in a basic set in an organism is called its monoploid number (x). Most organisms have two sets, diploid (2x). Anything more than 2 sets is called **polyploidy**, you can name them more precisely as triploid (3x), tetraploid (4x), pentaploid (5x).....

Research to add information to the table below.

	Natural	Artificial
Monoploids	Male bees, wasps and ants	Plant breeding
	Created pathogenically (from an unfertilised egg). Will not produce gametes normally. So, to fertilise the cells do not undergo meiosis at all.	Using a pollen grain (monoploid), induced to become an embroyonic mass which can be made into a plantlet on agar. This will not have any hidden recessives etc. If the plant is what you want, induce polyplodyism gives you the diploid (fertile) plant. All genes homozygous at each locus.

Colchicine is a drug that causes **spindle fibre failure**, **i.e. non-disjunction in the cell**.

.....

Polyploids – Plant examples banana, potato, swede, strawberry.....

What effects do polyploidy give to the plants generally bigger; bigger leaves, stems, fruits, seeds, greater number of seeds...

Why is polyploidy rare in animals ? there are problems with forming gametes and the animal physiology makes it less likely to survive changes in expression levels of proteins during development.

<u>Evolution</u> = the changes in organisms over generations as a result of genomic variations.

Extended Response;

Describe, with examples, the important of polyploidy on;

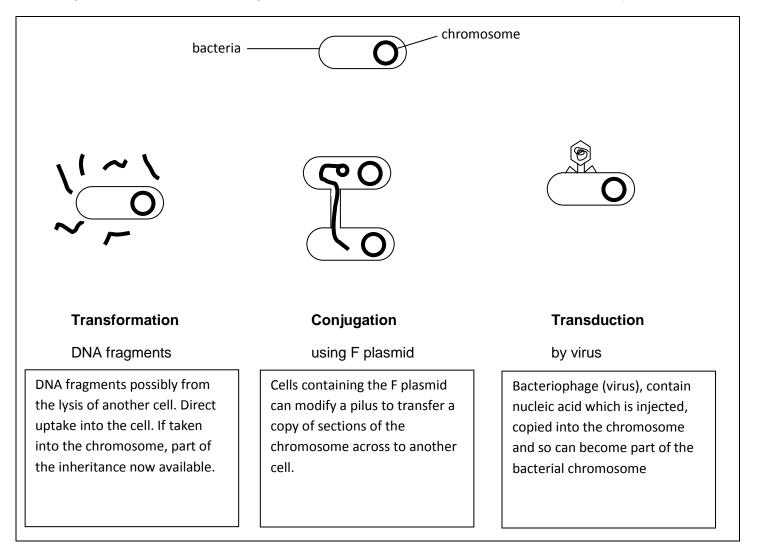
- a) Crop plants (5)
- b) Evolution (5)

Gene transfer

Transfer from parents to offspring, from one generation to the next, hence vertical gene transfer.

Bacteria can transfer DNA material within a population of the same generation, hence <u>horizontal gene</u> <u>transfer</u>.

The diagram below shows forms of genetic transfer in bacteria. Add as much information as you can.



What form of prokaryotes transmission shown above could also be used for horizontal transfer in eukaryotes? Explain your answer

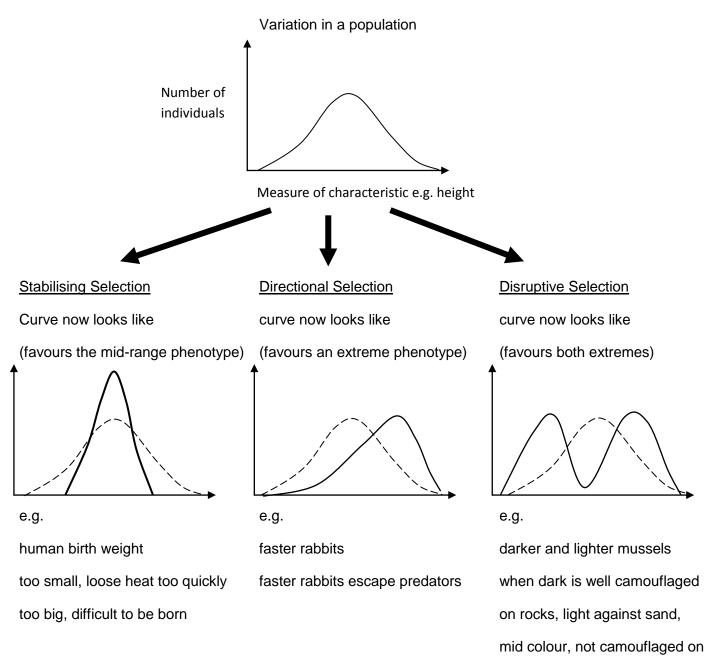
Conjuction – information is passed between individuals of the same generation, not down to offspring.

Technically transformation could also be within one generation if, one cell undergoes lysis and another picks up the fragments. Similar argument for transformation.

Selection

Natural selection is the non-random increase in frequency of DNA sequences that increase survival.

Sexual selection is an increase in successful reproduction.



either

Genetic drift

Definition changes in the gene frequency of alleles within a particular population due to chance loss of alleles due to death of individuals / lack of reproduction.

.....

Founder effects

Changes in the gene frequency can be found when a population goes through a bottleneck.

Research to find the following facts about Northern Elephant Seals

Northern Elephant seals went through a bottleneck in the 1890s

This was due to **overhunting**

The population reduced to around 20 indivduals

All of the alleles present in the current population were present in these individuals (except from a few changes caused by **mutation**.)

The present population is around 30,000

Find some more examples of founder effects in populations

Afrikaner population in S. Africa

The seven daughters of Eve – mitochondrial Eve

Island populations after large scale impacts

Neutral effects

Neutral mutations are liable to genetic drift because they are not liable to natural selection.

Explain this statement

Mutations that are not involved in selective pressures, will be subject to random effects. i.e. they have no impact on the selection itself instead they will increase or decrease in a totally random way Genetic drift is the random increase/decrease in small populations.

.....

Speciation

Species = a group of individuals that can interbreed to produce fertile offspring.

Speciation = the formation of a new species

Stages in Speciation (basics covered in N5) – add notes for each stage

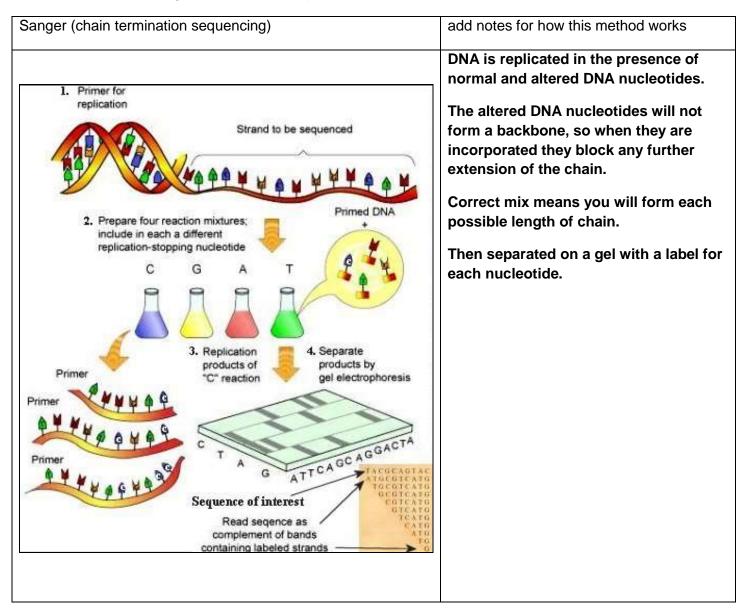
Step	Description / Notes	
1 (split)	A population is split into two groups	
2 (isolate)	By barrier;	
	Geographical e.g. mountains or sea	
	Ecological e.g altitude, humidity barrier, pH	
	Reproductive e.g breeding season moves out of synch	
3 (variation)	Created by mutation in the population	
4 (pressure)	Different selective pressures favour different phenotypes/ alleles	
5 (change)	Selection (and genetic drift) alters the gene pool	
6	Barriers removed.	
7 (speciation)	The two populations can no longer produce fertile offspring = new species	

How is <u>Sympatric Speciation</u> different from <u>Allopatric Speciation</u> sympatric happens in the same habitat, allopatric occurs in different habitats. i.e. sympatric will be due to reproductive barrier or due to disruptive selection over a long time. Allopatric will involve a geographical barrier.

.....

Sequencing

Genes and even whole genomes can be sequenced.



Extra; research how shot-gun sequencing works

Describe the part that computers and statistical analysis play in genome analysis

Computers can be used for comparisons on data within and between species, including looking for start/stop or coding sequences. Or mutations

Evidence for evolution

Describe what is meant by each of the following and explain how they show evidence for evolution.

	Description	Evidence
Fossil Record	Preserved remains or imprints of organisms (animals / plants / micro-organisms)	Since rocks can be dated you have a progressive changes that can be tracked and points in time when species are removed or appear.
Phylogenetics	Study of genetic relationships using sequences to determine how closely species are related	By looking at common sequences you can group species together. You can how groups of species come from common ancestors.
Molecular clocks	Uses the rate of change in molecules to work out the time it has been between divergence events.	Shows closely species are related and how long it has been since common ancestors were present.

What reasons are there for sequencing whole genomes you can find all coding sequences, gives insight into conserved areas. Comparative mapping in species and between species. Find disease alleles. Find information on expression sequences.

Personal genomics and health

Pharmacogenetics genetic differences which cause individuals to react differently to pharmaceuticals (drugs)

Give one reason why you think research in personal genomics will progress in the future

ARA; technology allows it to be more likely, there are more individuals that can access the information/ have the finances to drive personal genomics. Risk based insurance. People living longer....

Give one ethical issue with the use of personal genomics and state your opinion regarding this issue.

ARA; insurance companies getting data on risk affecting life insurance policies. Employers getting data on health risks including e.g. risk associated with heart attacks, some mental health....