Unit 1

DNA and the Genome

National 5 Knowledge

Learners should have a clear understanding of the following areas of content from their previous learning:

*Cell division (mitosis) and chromosomes
*Base sequence and base pairing of DNA
*Function of proteins
*Evolution by natural selection
*Species
*Classification of life
*Cell ultra-structure and function

Key Area 1

- (a) Structure of DNA
- (b) Organisation of DNA

(a) Structure of DNA

Vocabulary:

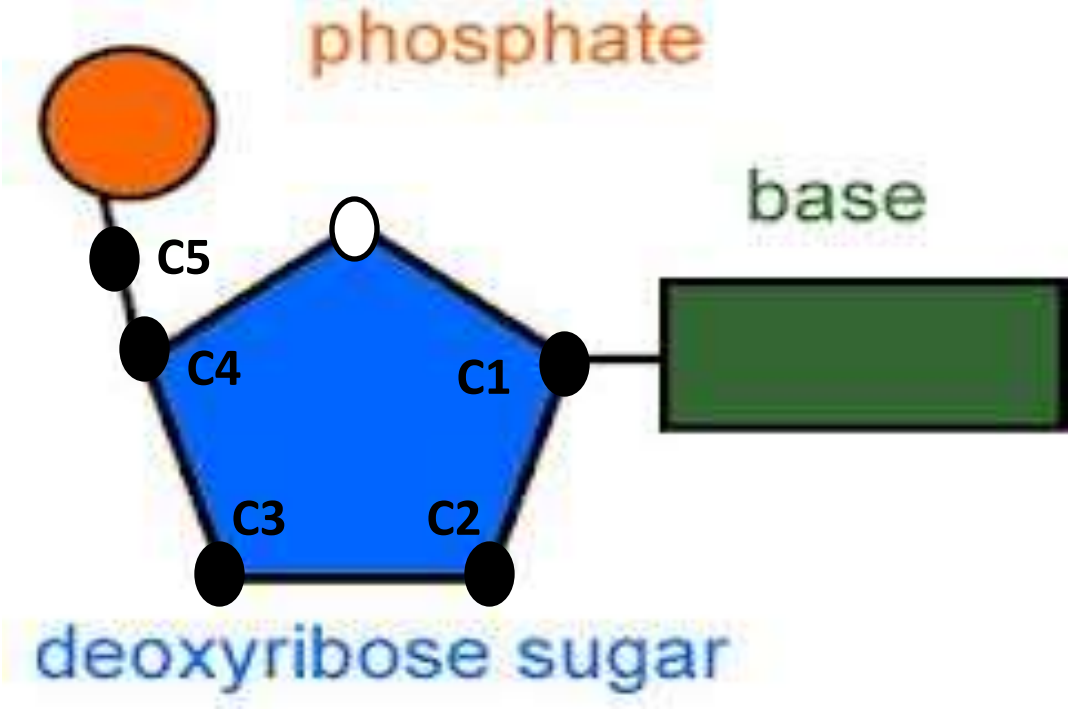
- Genotype
- Nucleotide
- Sugar-phosphate backbone
- Complementary base pairing
- Hydrogen bonds
- Double stranded
- Antiparallel
- 3’ and 5’ end
- Double Helix structure

Genotype

- The genotype is the genetic material of an organism. It is determined by the sequence of bases along the DNA molecule.

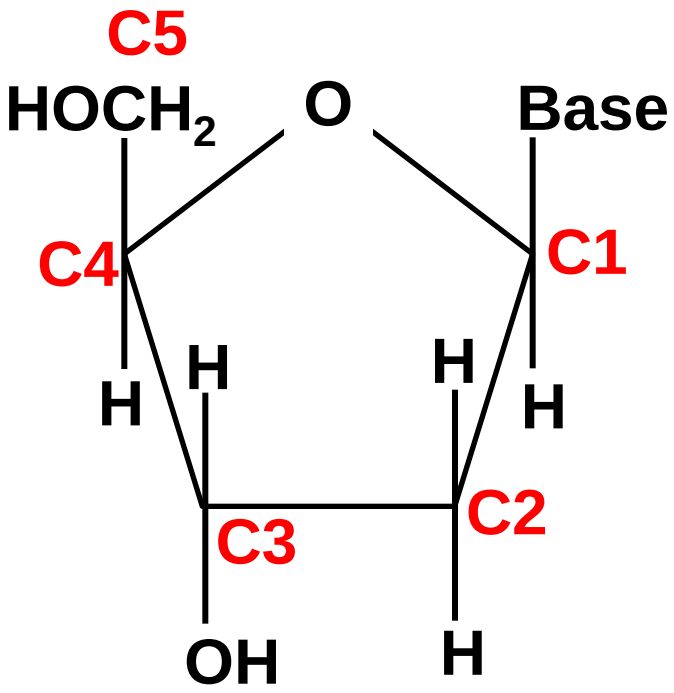
Structure of DNA

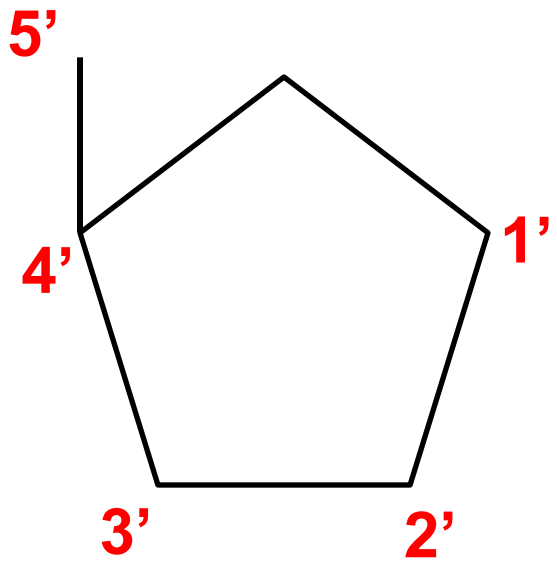
- DNA stands for DeoxyriboNucleic Acid.
- It is a double stranded molecule
- Each strand is composed of **repeating** chemical units called nucleotides.
- Each nucleotide is made of a deoxyribose sugar(5C), a phosphate group and a base.



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

another way of showing this is



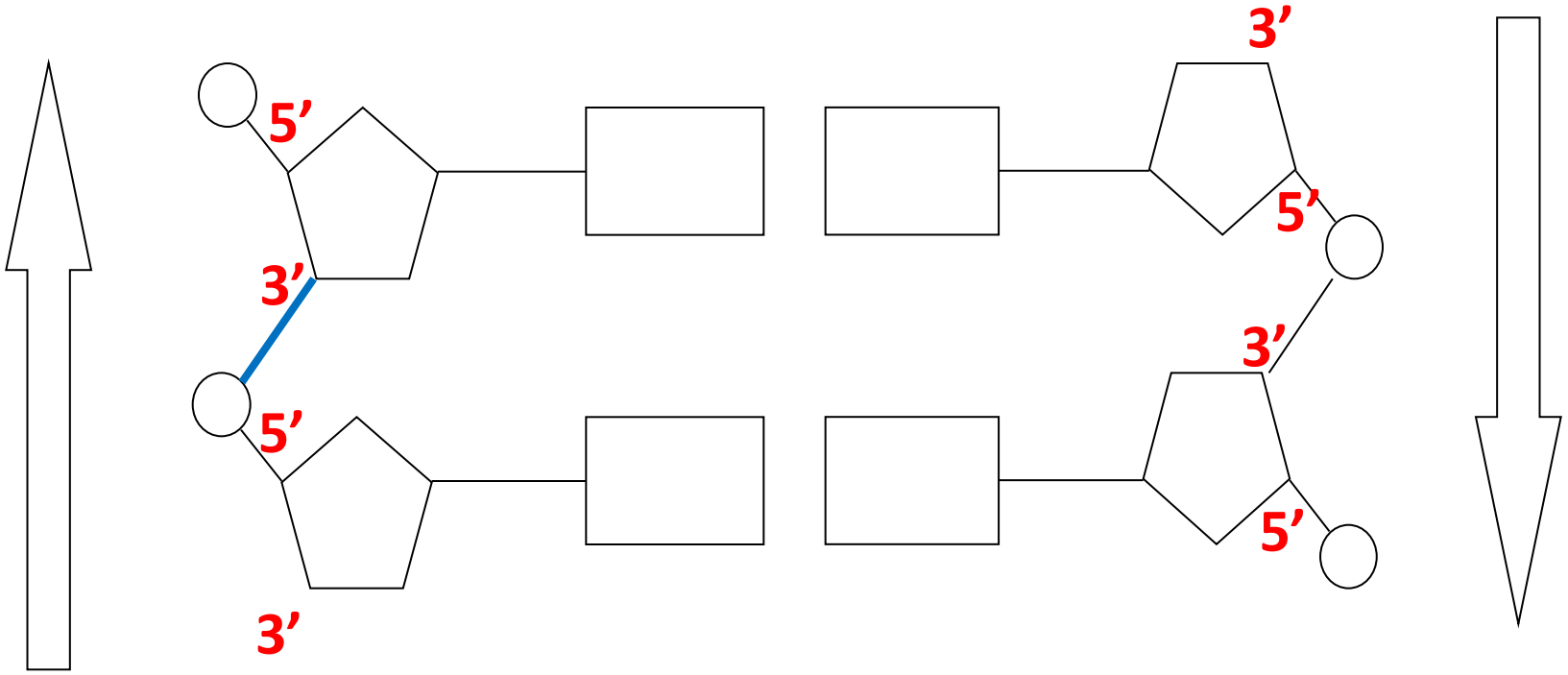


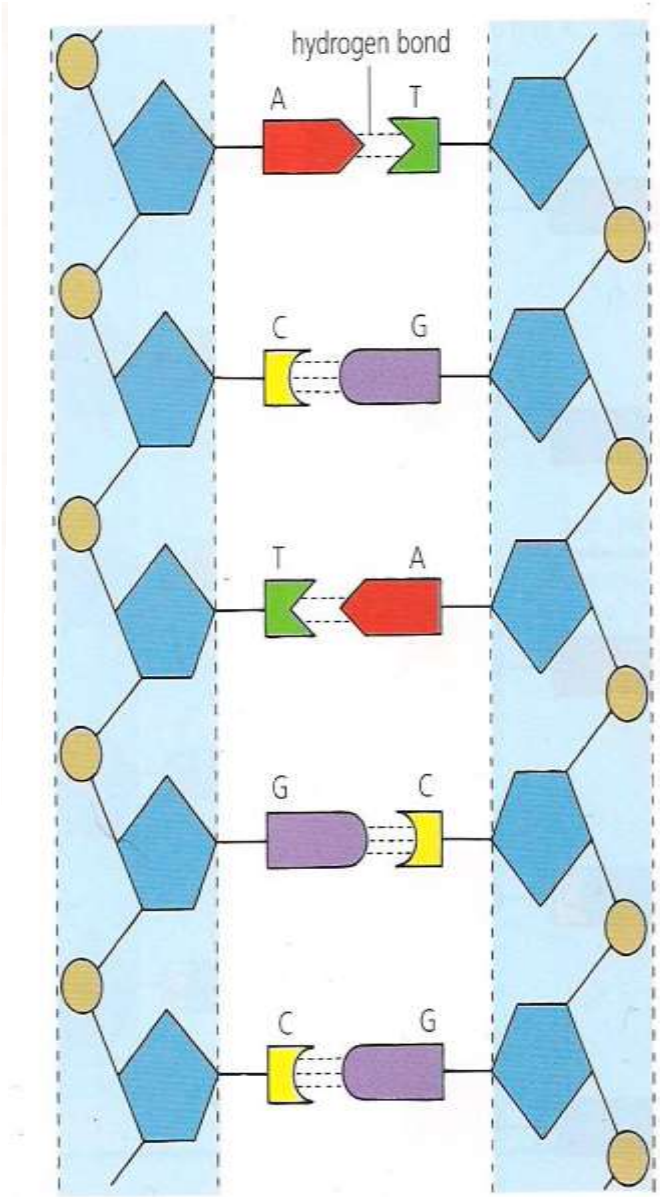
5′ pronounced "5 prime"

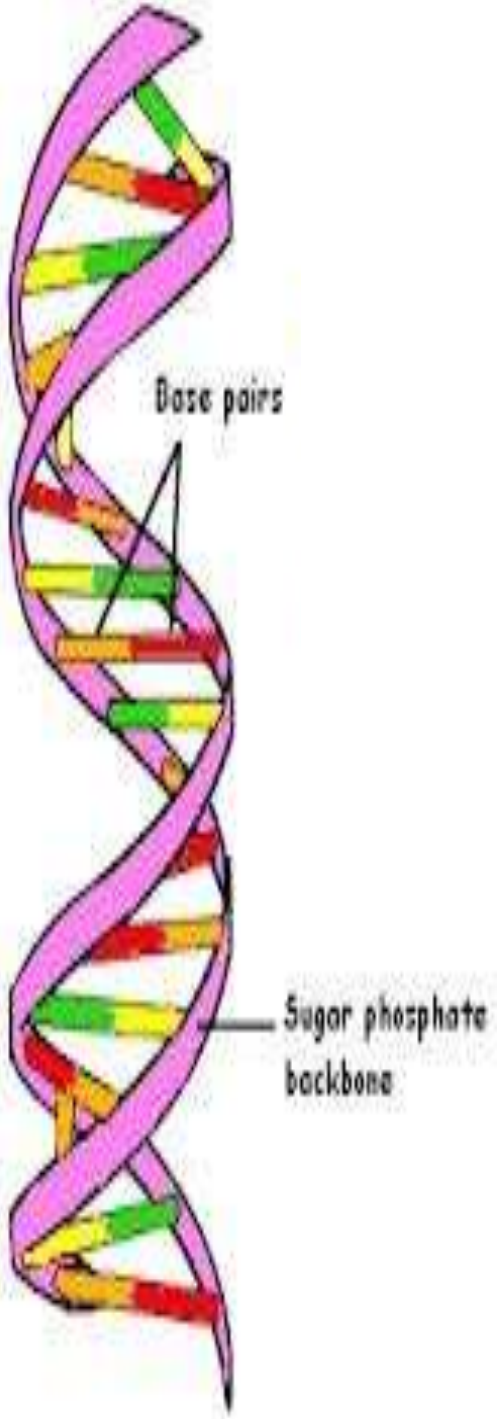
Activity

- Draw 4 repeating nucleotides (one underneath each other)
- How are they attached to form a strand of DNA?
- This produces a sugar-phosphate backbone with the bases attached
- Now turn your page upside down and draw 4 more nucleotides with adjacent bases beside each other...I'll demo!
- How are the 2 strands attached to form the double helix structure?

Be able to state that:
there is a deoxyribose sugar at the 3’ end and a phosphate at the 5’ end




hydrogen bond
A
T
C
G
T
A
G
C
C
G


Base pairs
Sugar phosphate backbone

Complementary Base Pairs

- Adenine ALWAYS bonds with thymine.
- Cytosine ALWAYS bonds with guanine.

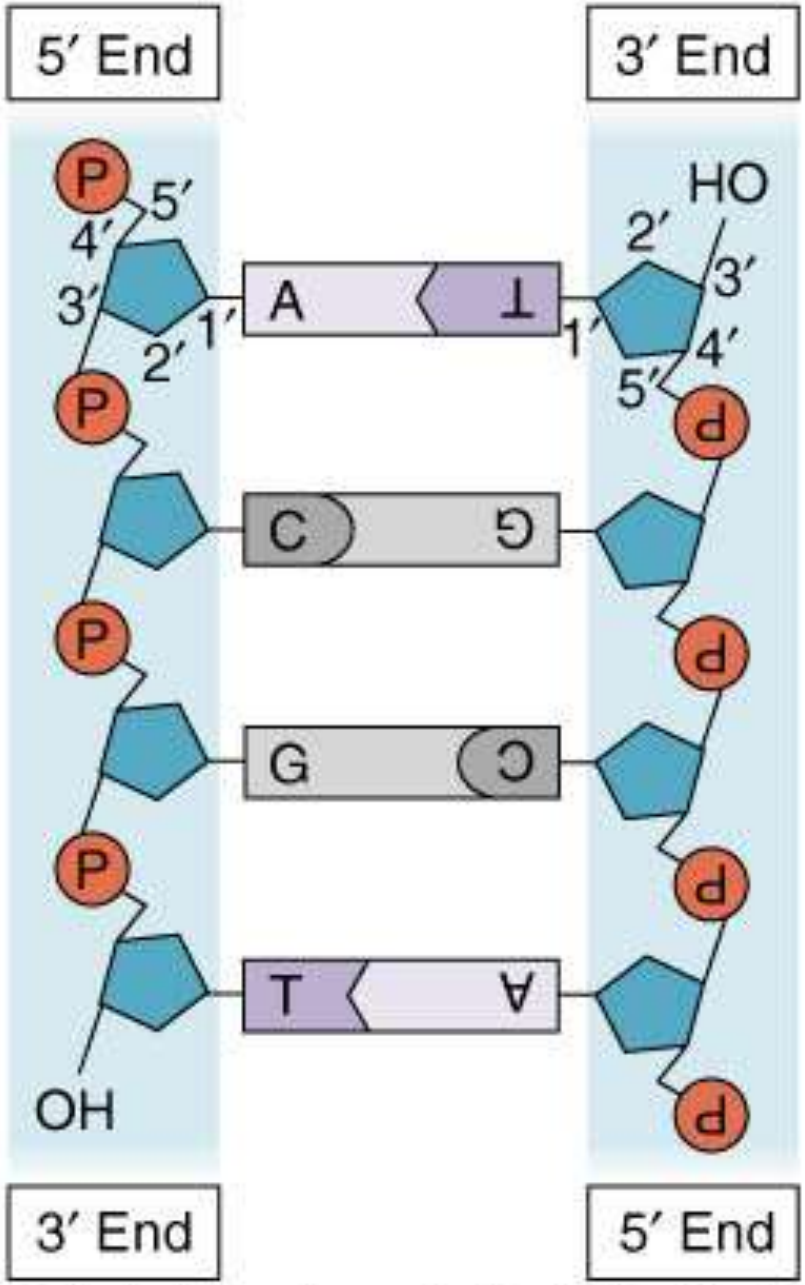
- A-T and C-G are called base pairs.

Antiparallel Strands

- Go back to your drawing and i'll explain a bit more.

- The 2 strands run in opposite directions. One from 3' to 5' end and the other from the 5' to 3' end.

5P



Double Helix Structure

- The double helix shape of DNA forms due to the base pairs aligning

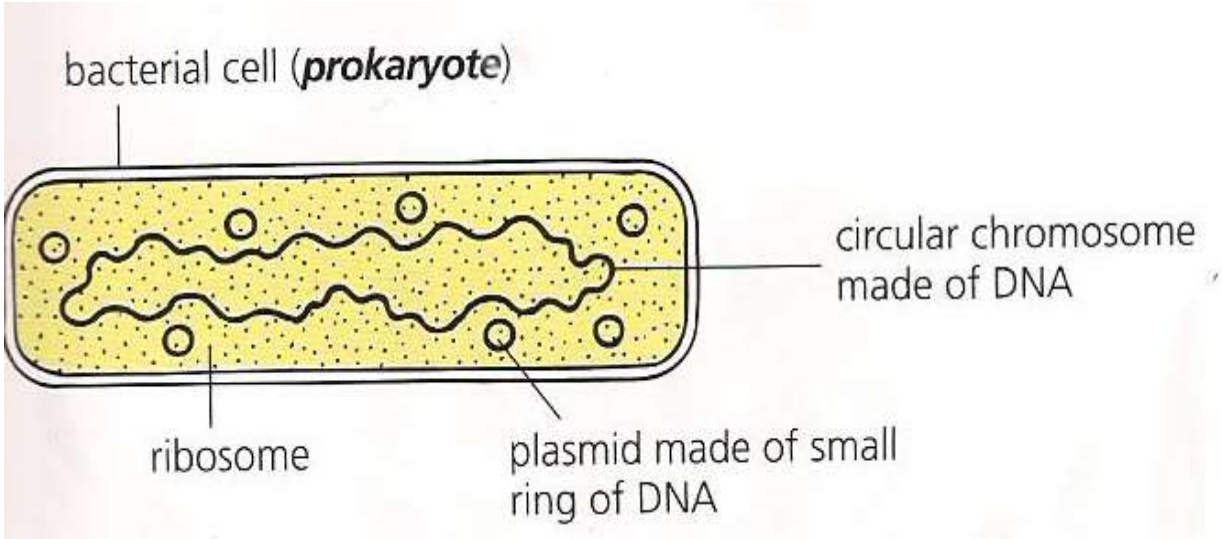
(b) Organisation of DNA

- **<u>Vocabulary</u>**:

- Prokaryote
- Circular chromosome
- Plasmid
- Eukaryote
- Linear chromosomes

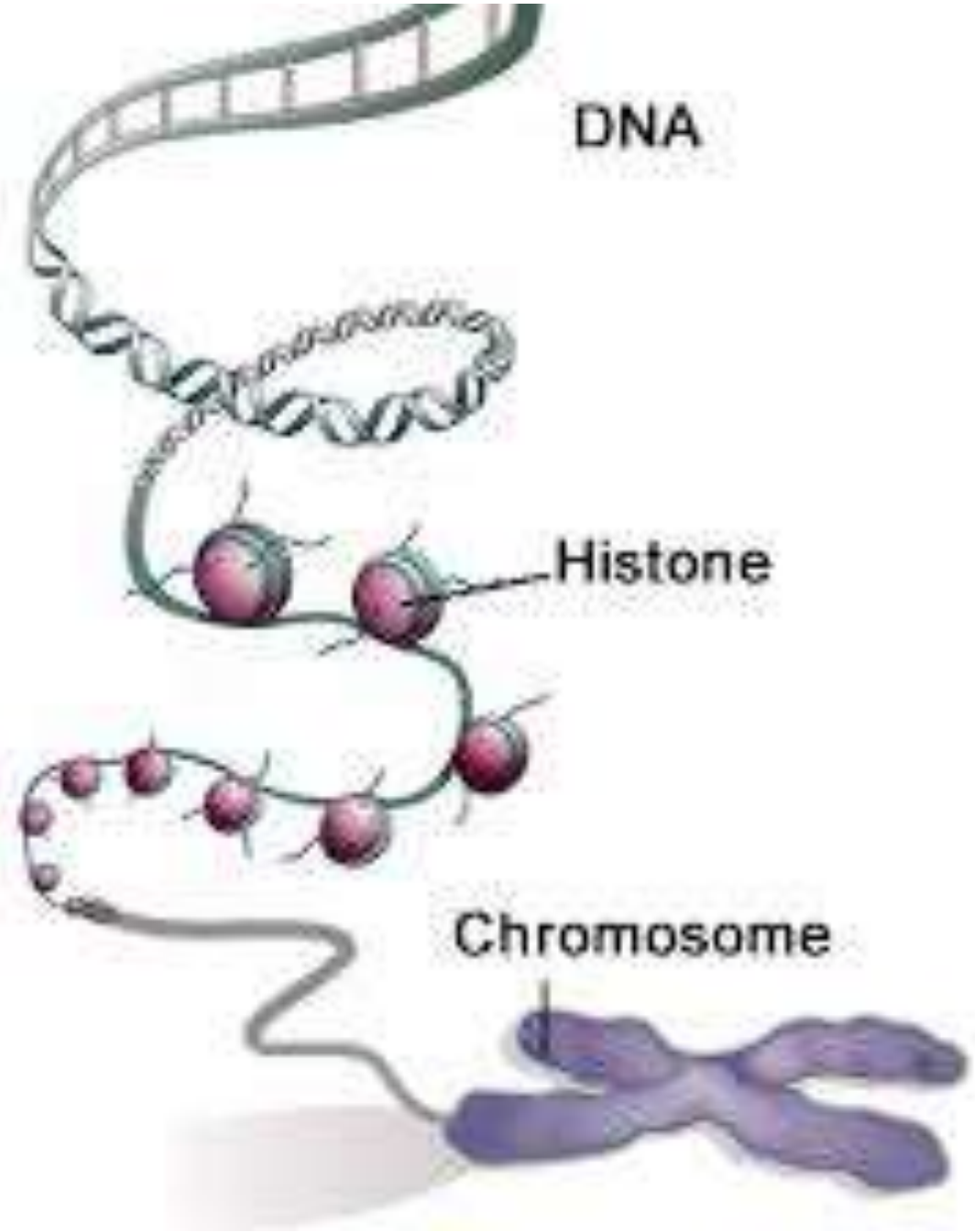
Prokaryote

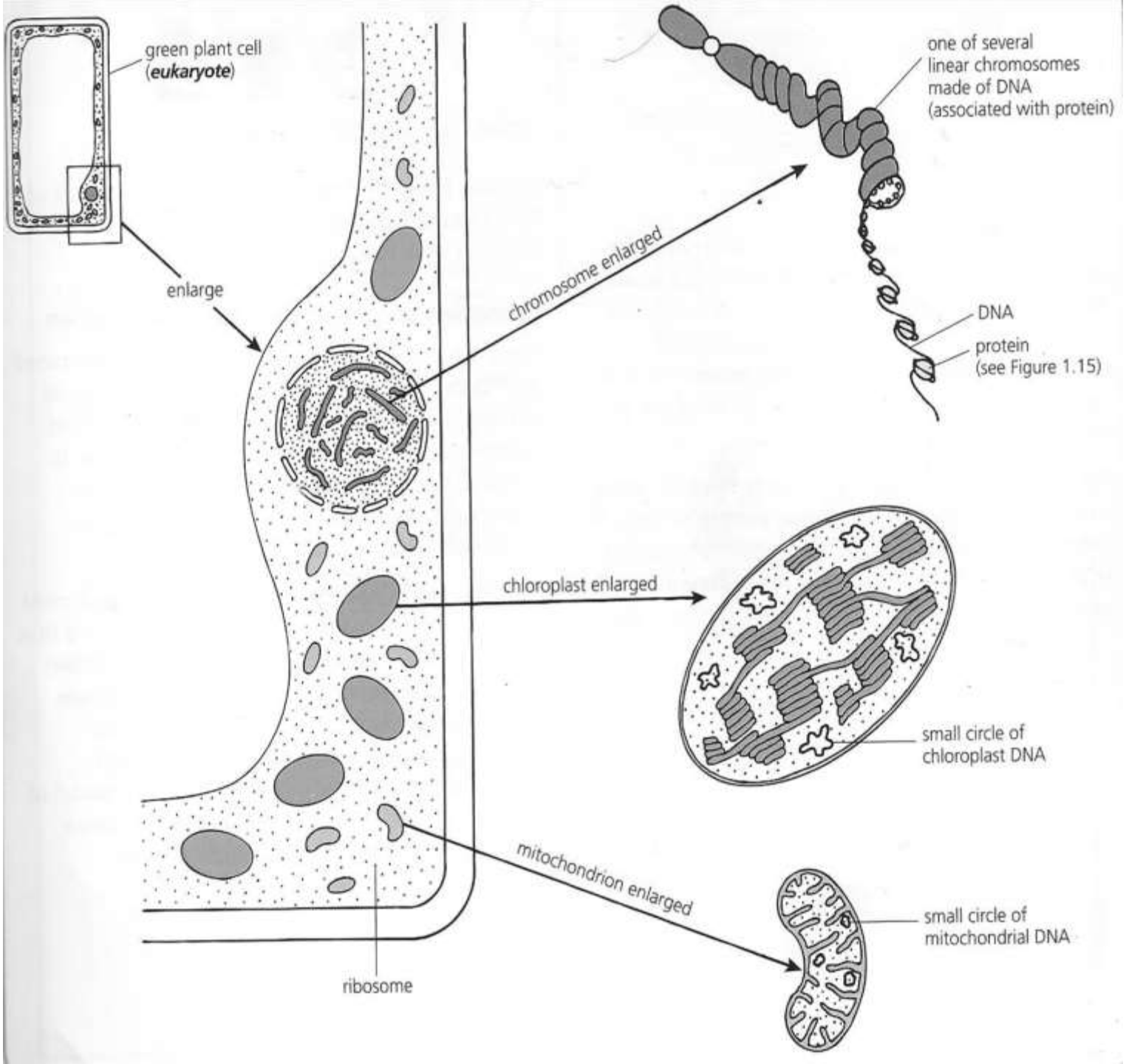
- Organisms with a single cell which do not have a nucleus.
- The DNA in organised in <u>circular chromosomes</u>.
- They also have smaller rings of DNA called <u>plasmids</u>.
- E.g. Bacteria



Eukaryote

- Eukaryotic cells have a nucleus. The DNA in the nucleus is organised as linear chromosomes.
- The DNA is tightly coiled around bundles of protein (histones).
- These cells can also have small circular DNA in the mitochondria and chloroplasts.
- E.g. Animals, plants and yeasts.


DNA
Histone
Chromosome


green plant cell
(eukaryote)
enlarge
one of several
linear chromosomes
made of DNA
(associated with protein)
chromosome enlarged
DNA
protein
(see Figure 1.15)
chloroplast enlarged
small circle of
chloroplast DNA
mitochondrion enlarged
small circle of
mitochondrial DNA
ribosome

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

Key Area 2

- Replication of DNA

Replication of DNA

- **<u>Vocabulary</u>**:

- Semi-conservative replication
- ATP
- Enzymes (Helicase, DNA Polymerase and Ligase)
- Primers
- Parental DNA strand
- Daughter DNA strand
- Leading strand
- Lagging strand
- Replication fork
- PCR
- Thermal cycler
- Amplification
- In Vitro

Replication of DNA

Semi-conservative replication means

Half of the new DNA comes from the old DNA (i.e you kept (conserved) half (semi) of the original.

Requirements for DNA replication	Reason you need it
1. DNA template	Provides the code to copy
2. Supply of DNA nucleotides	To produce the new strand of DNA
3. Supply of energy (ATP)	To provide the enzymes with energy to; unwind the DNA, pair up the new nucleotides, join up the backbone, rewind the DNA spiral
4.Enzymes a. Helicase b. DNA polymerase c. Ligase	a) Unwind the DNA (and hold replication fork open) b) Pair up the free nucleotides c) Join up the Okazaki fragments
5. Primers for DNA polymerase	To allow the attachment of DNA polymerase

DNA replication

Okazaki fragments =

the DNA fragments that form on the lagging strand

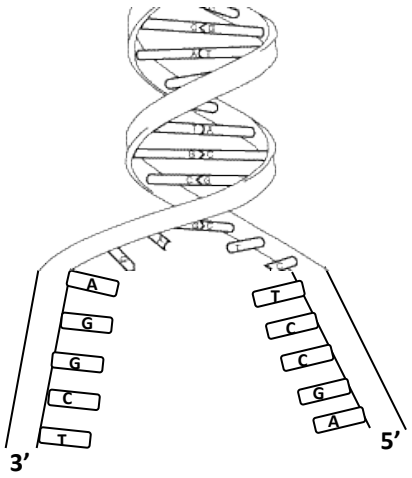
Replication fork =

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.

Complete the following diagrams 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



DNA untwists and 'unzips. Requires enzymes (helicase) and energy (ATP). Gives you two ends and a replication fork.

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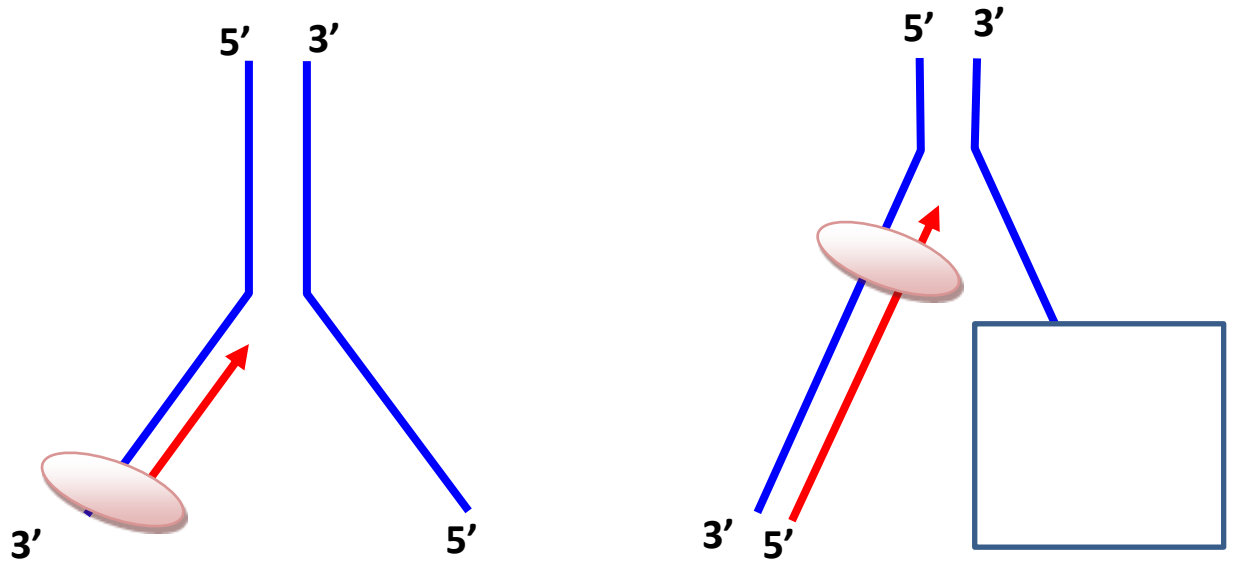
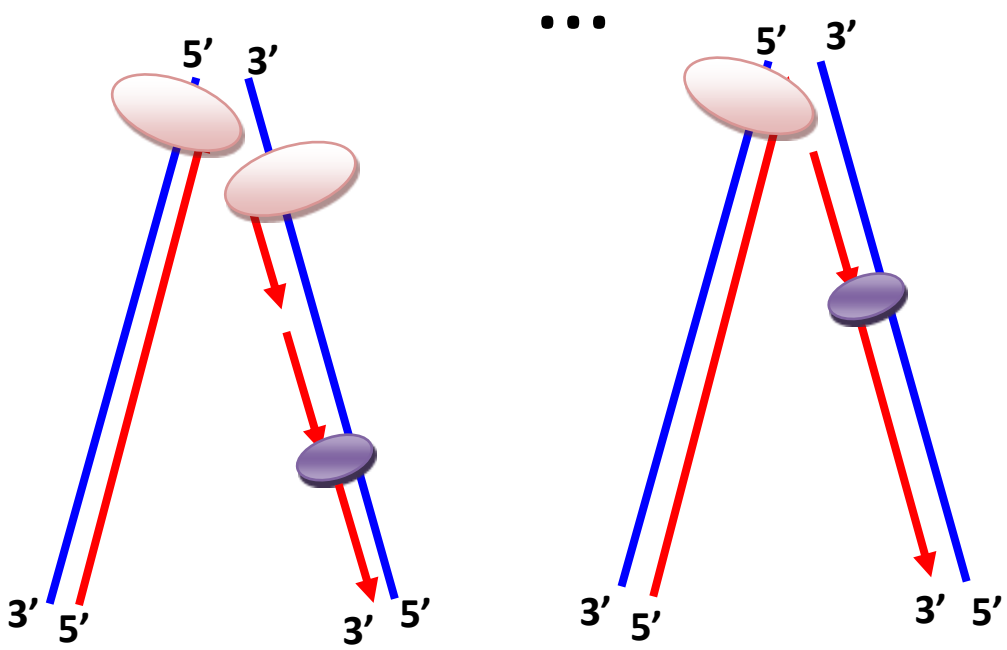


DNA polymerase can only read 3'→5', so attaches to the 3' end creating the leading strand.
This will replicate continuously.
Adding complementary nucleotides to the chain.
As the replication fork moves DNA polymerase can attach and read back down from the 3' end to the 5' end on the other strand (lagging strand)
The DNA polymerase drops off the end

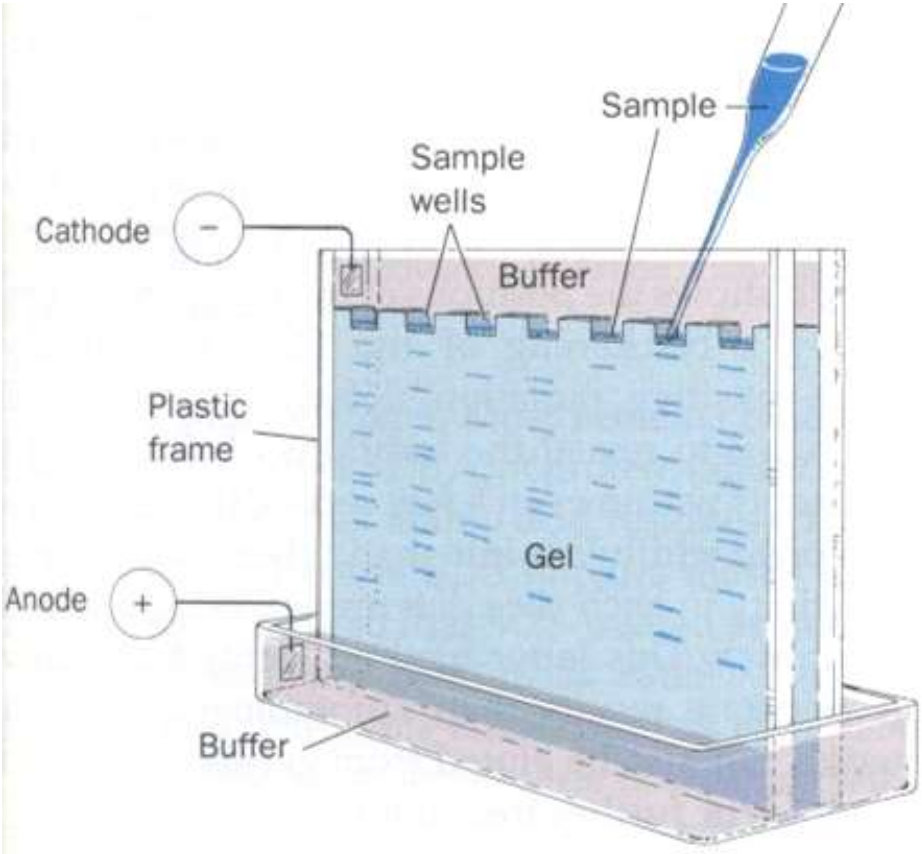


As more of the lagging strand becomes available more fragments are formed. (Okazaki fragments)
These fragments are joined together by the enzyme ligase creating a continuous strand.
At the end you have two identical strands, each containing one strand from the original molecule = Semiconservative replication

Importance

- Allows specific proteins to be synthesised

Gel electrophoresis – separating DNA



Explain how gel electrophoresis works

Agarose forms a network that the strands of DNA can move through.
DNA is negatively charged. So will move away from the negative terminal.
The smaller the DNA fragment the faster/further it moves.

Polymerase Chain Reaction (PCR) –

Using DNA replication to **amplify** a **specific** section of DNA

Stage	Temperature (°C)	What is happening
1	92	DNA strands at the bases are denatured (pull apart)
2	55	Primers bind to target sequence
3	72	Heat-tolerant DNA polymerase activity

PCR

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

PCR

In Stage 3 Why is *Taq* polymerase used?

Originally extracted from a thermophilic bacteria – will not be denatured by the high temp.

Applications of PCR

- DNA sequencing
- Genetic mapping
- Forensics
- Parental testing
- Sex determination in pre-natal cells
- Classification of species into taxonomic groups
- Screening for genetic disorders