

DNA structure:

Nitrogenous bases can be:

- Purines – double carbon/nitrogen ring (A and G)
- Pyrimidines – single carbon/nitrogen ring (T, C and U)
- Nucleoside → base + sugar
- Nucleotide → base + sugar + phosphate group

Primary

- Sugar phosphate backbone
- Linear sequence of bases in hydrophobic single strand
- Strand has orientation (5' to 3')

Secondary

- Nitrogenous bases form hydrogen bonds (A-T forms 2, G-C forms 3)

Tertiary

- Double helix
- Major groove gives better access to DNA
- Antiparallel
- Phosphodiester bond (3 OH' and 5' phosphate)

RNA

- Extra OH' group at 2'

Functions vs Structure of DNA

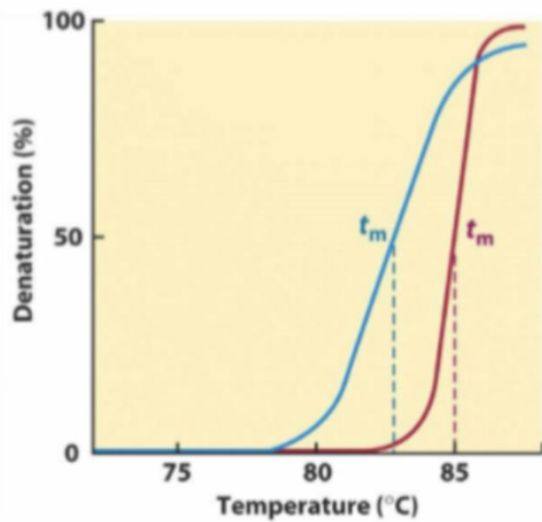
1. **Storage of the genetic information: sequences of bases which order is maintained by covalent phosphoribose backbone and made of four different nucleotides**
2. **must be precisely copied and must be read to allow gene expression: base complementarity allows coping and reading of DNA and weak bonds between strands.**
3. **Susceptible to mutations: mutations affect nucleic bases.**

Major Species of RNA:

- **mRNA:** messenger RNA, codon codes for amino acids → polypeptide
- **tRNA:** transfer RNA carry specific amino acids to ribosome
- **rRNA:** make up small and large ribosomal subunits.
- **snRNA:** splicing of premRNA in nuclear processes

Structural features of DNA:

- Double stranded helix
- Hydrogen bonding maintains helix
- Complimentary base pairing
- Long length and thin diameter
- Acidity due to negatively charged phosphates



Complexity:

- **Steepness of curve= complexity of DNA**
- Steeper curve= less complex
- Red is less complex than blue

Formulas

$$T_m = 69.4 + 0.41(\%G+C) - 650/N$$

Factors that impact Tm:

- **GC content:** higher GC content means it takes more energy (higher T_m) to melt DNA of the same length
- **Size:** shorter DNA denatures quickly. Increases the steepness of the curve and increased length increases T_m
- **Complexity:** complexity increases T_m
- **pH and ionic strength:** higher pH means DNA is less stable, decreases T_m and easier to separate the 2 strands
- (denatures easily) and higher salts means it stabilises the ds molecule and increases t_m.

Gel electrophoresis:

- Move towards positive anode because negative phosphate groups
- Smaller fragments move faster/go further
- Supercoiled goes fastest, then linear, then circular
- Only linear DNA can be used to estimate the size of DNA fragments

- Semi-Conservative-Each strand acts as a template then formation of a DNA strand occurs between the parent and daughter DNA strand
- Conservative- Original double helix of parental DNA is maintained
- Dispersive-Two strands of parental DNA totally disintegrate producing two new daughter cells completely unique of parental cell as parental DNA contains fragments of daughter DNA

DNA replication:

Function of enzymes in DNA replication:

Helicase

The enzyme responsible for separating double stranded DNA into single stranded DNA with aid of ATP(breaks hydrogen bonds) → localised denaturation of DNA at the replication fork.

SSB: The unwound DNA is stabilised by SSB. SSB binds tightly to the backbone and without covering the bases.

- Holds the DNA strands apart while they are being replicated.

DNA Primase

The enzyme responsible for synthesizing a short strand of DNA template, producing a primer for DNA synthesis.

RNA Primer

A short stretch of RNA synthesized on a DNA template. It is required by DNA polymerase to begin DNA synthesis.

DNA Polymerase

The enzyme responsible for synthesizing new DNA by joining nucleotides together on 3', using a DNA template as a guide.

- Binds to template strand
- Catalyses reaction
- Correct nucleotide has a higher affinity for polymerase (energy favourable)

Sliding clamp and clamp loader:

- Assist DNA polymerases by the use of the sliding clamp of DNA onto the polymerase.
- This process requires ATP
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Topoisomerase

The protein responsible for unwinding DNA in front of the replication fork → releases stress and ensures DNA rigid structure

DNA Ligase

The enzyme that joins the ends of two strands of DNA (the Okasaki fragments on the lagging strand) together with a covalent bond to make a continuous double DNA strand.