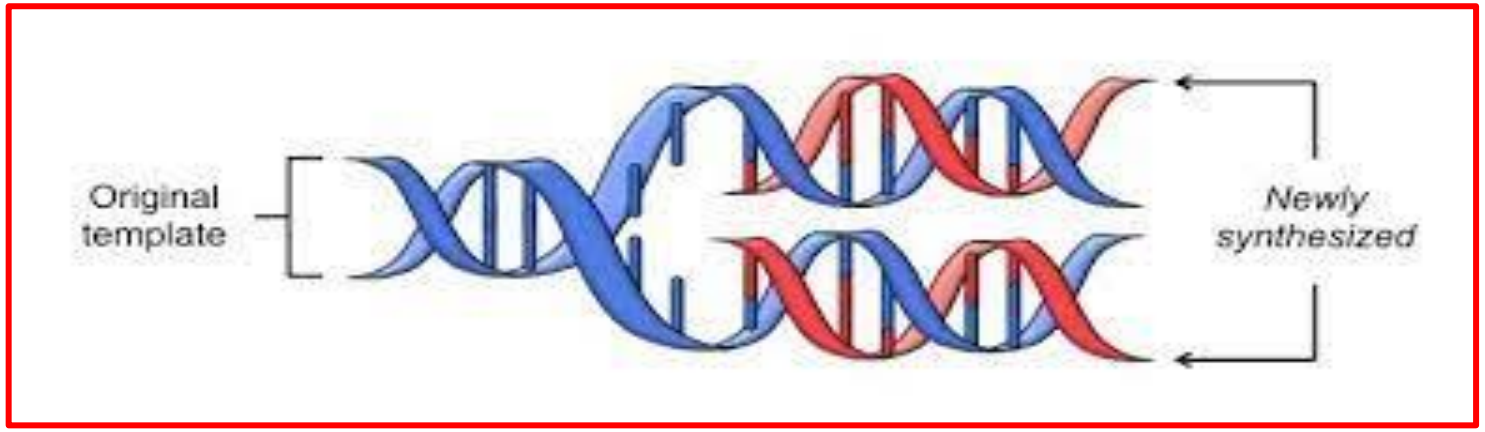

DNA Replication

DNA replication

- DNA replication is the process by which a double-stranded DNA molecule is copied to produce two identical DNA molecules. Replication is an essential process because, whenever a cell divides, the two new daughter cells must contain the same genetic information, or DNA, as the parent cell.

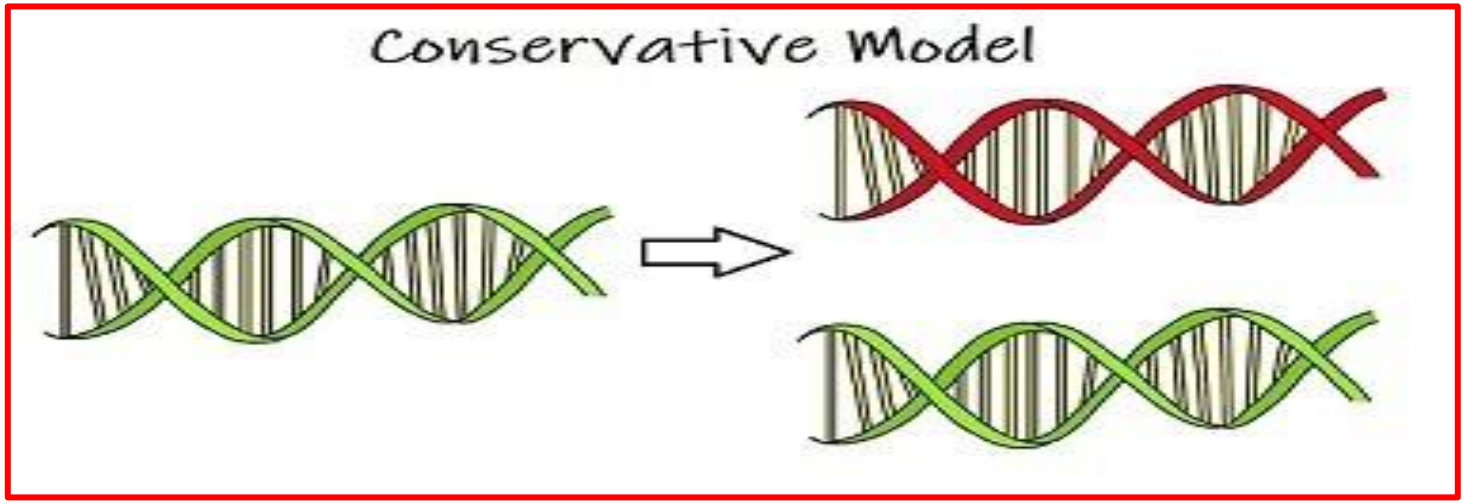
Models of Replication

- **1. Semiconservative DNA replication:** Semiconservative DNA replication: Two strands of the original DNA are separated, and each is duplicated by the synthesis of the complementary strand, thus producing two exact replica of the parent DNA. Thus, only one of the two strands of the original DNA is conserved. And this supported by Watson and Crick model for the DNA double helix.



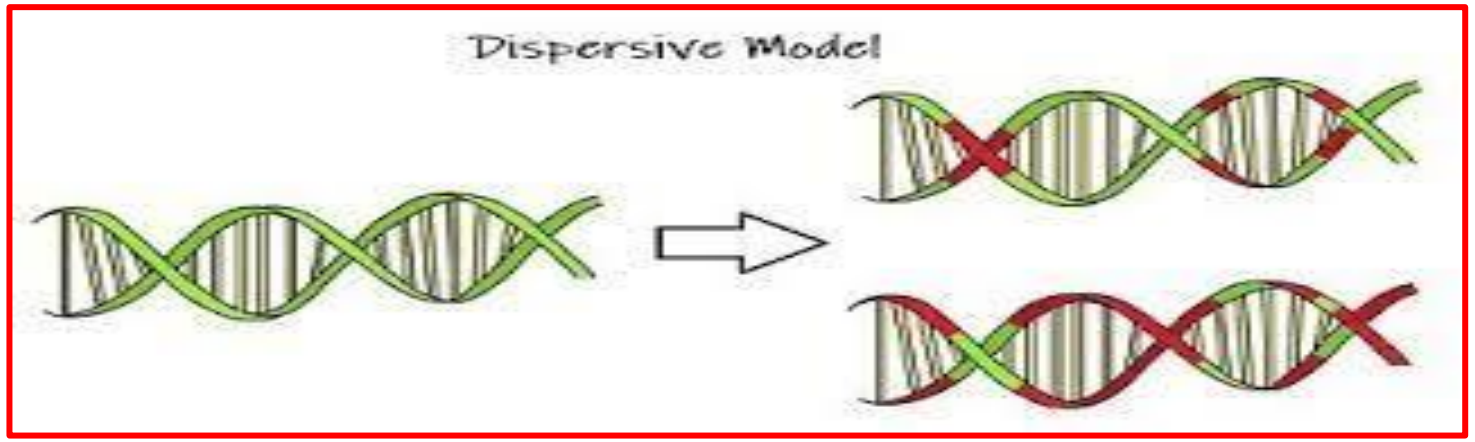
Conservative replication:

- **Conservative replication:** Two parental strands stay together, and two daughter strands stay together. The original helix is conserved.



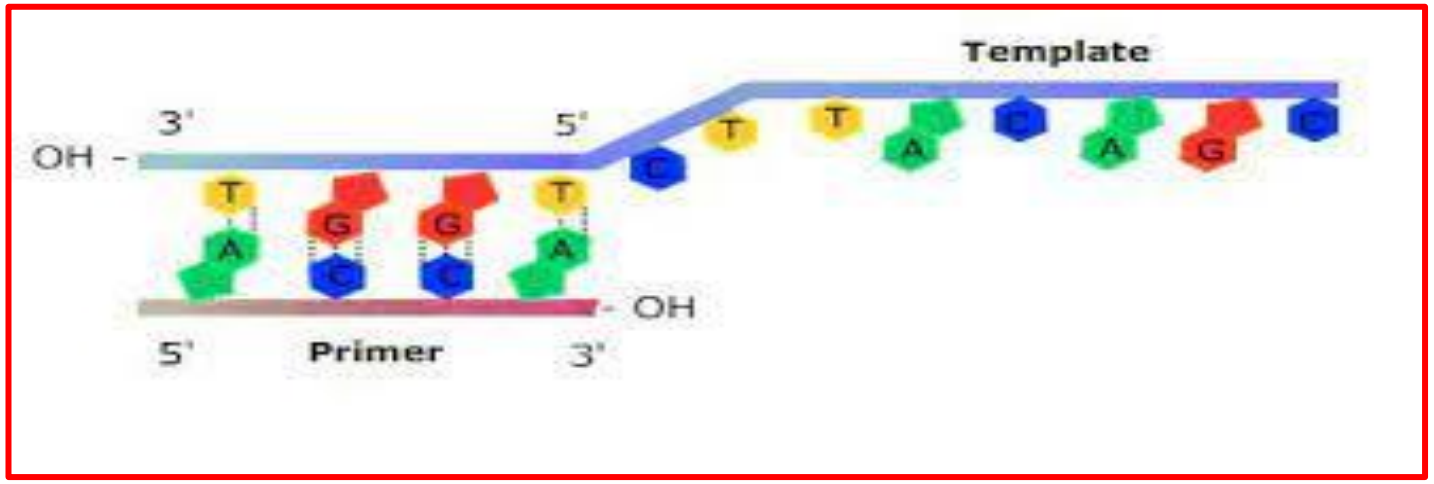
Dispersive replication

- **Dispersive replication** Parental and daughter material is mixed on each strand. It is the most complex of the three possibilities and is, therefore, considered to be least likely to occur.



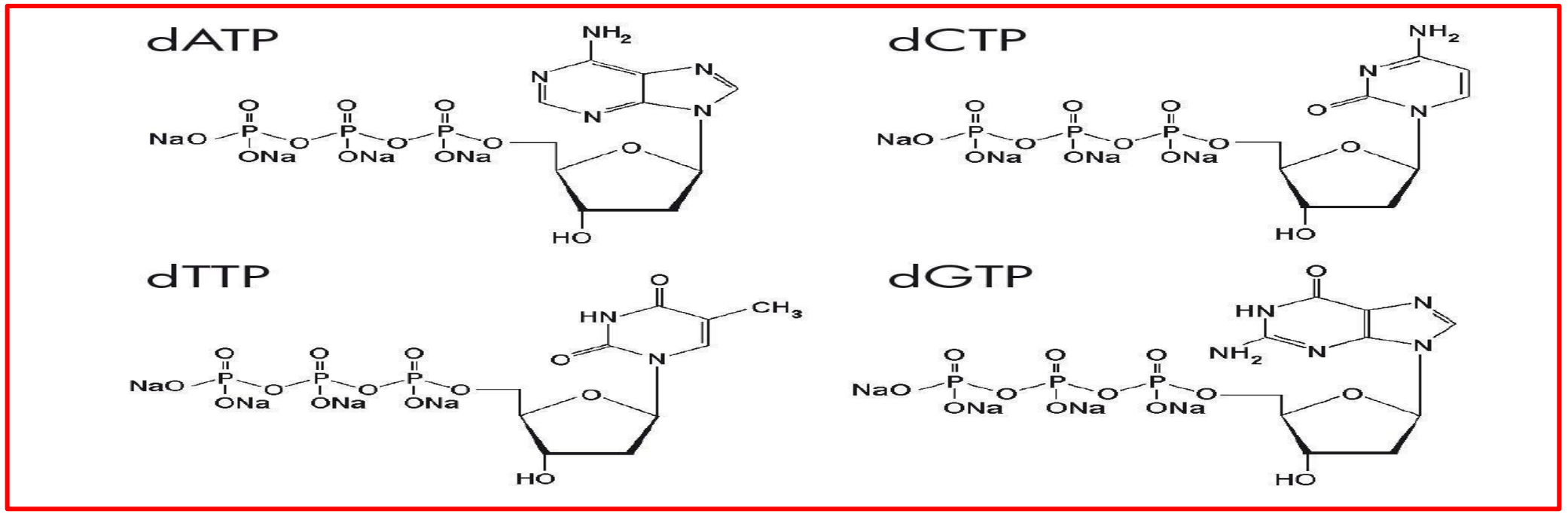
Requirements of DNA replication

- There are four basic components required to initiate and propagate DNA synthesis. They are: substrates, template, primer and enzymes.
- **1. Template**
- The nucleotide that is to be incorporated into the growing DNA chain is selected by base pairing with the template strand of the DNA. The template is the DNA strand that is copied into a complementary strand of DNA.

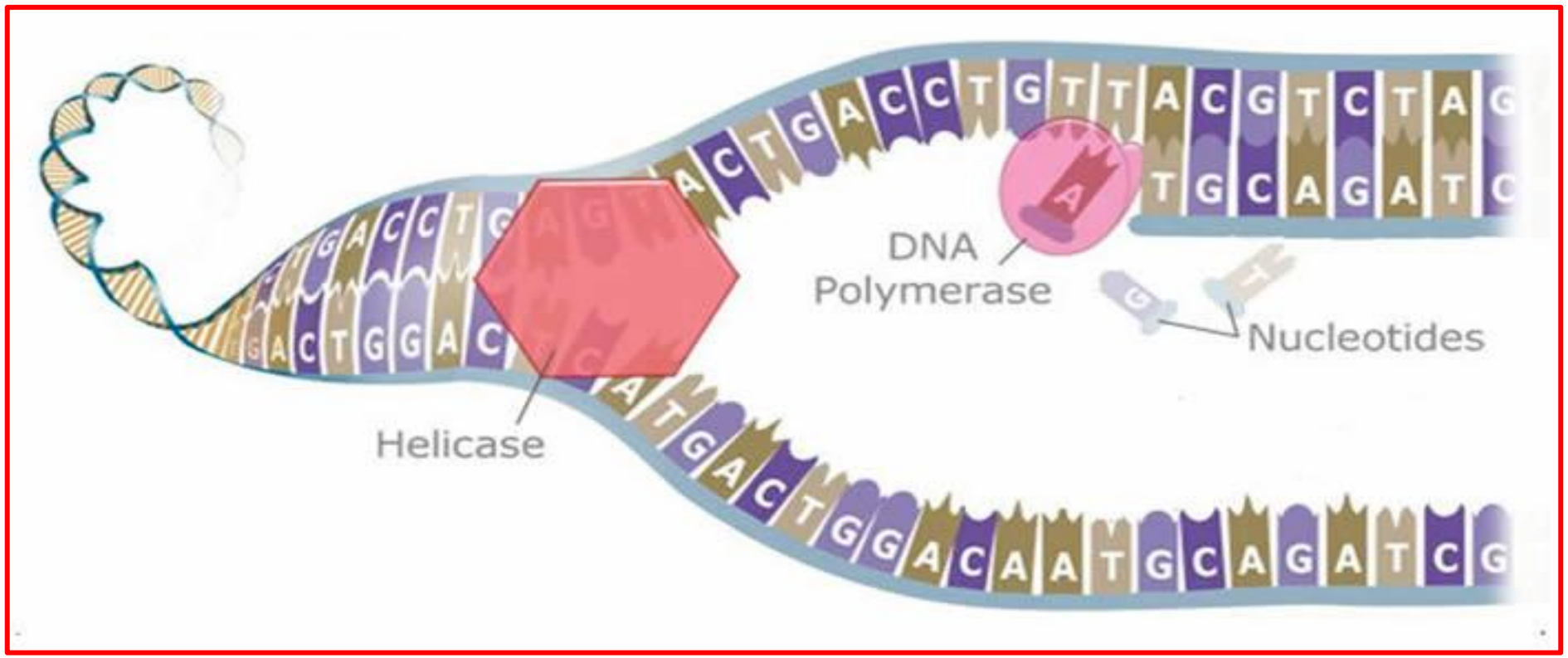


2. Substrates

- Four deoxyribonucleotide triphosphates (dNTP's) are required for DNA synthesis (note the only difference between deoxyribonucleotides and ribonucleotides is the absence of an OH group at position 2' on the ribose ring). These are dATP, dGTP, dTTP and dCTP. The high energy phosphate bond between the a and b phosphates is cleaved and the deoxynucleotide monophosphate is incorporated into the new DNA strand.

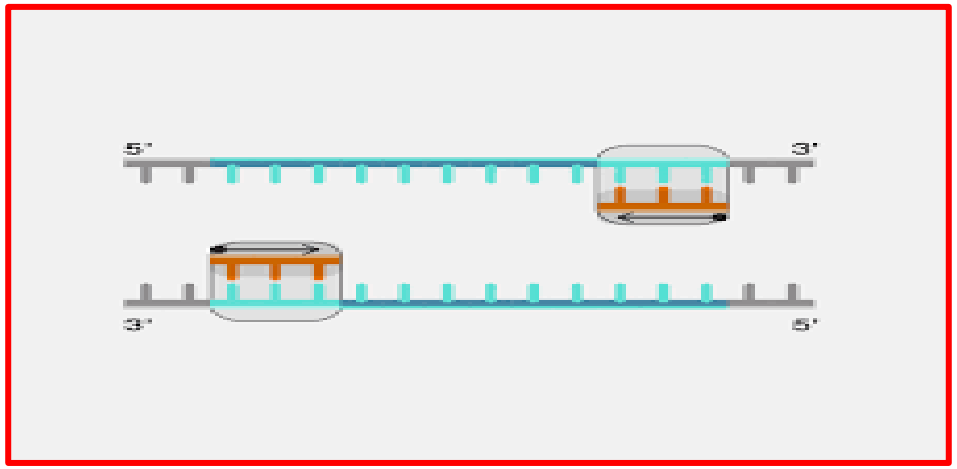


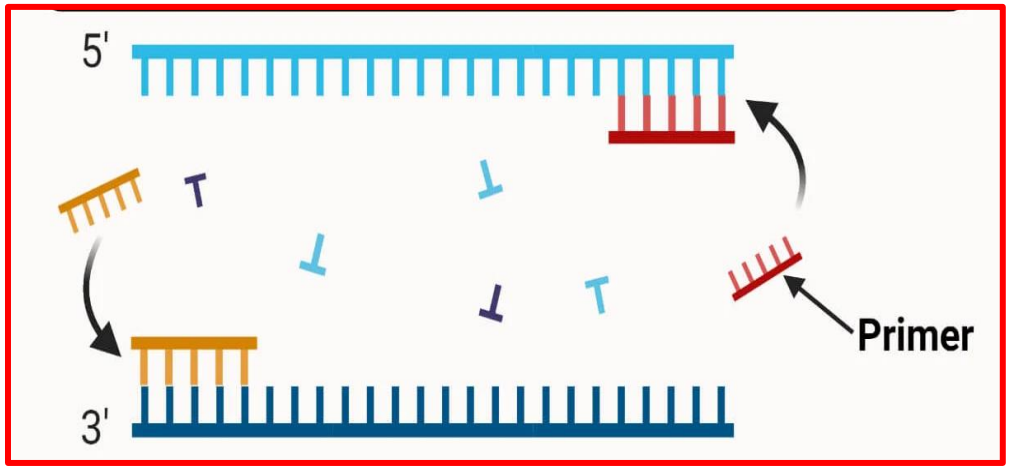
- Ribonucleoside triphosphates (NTP's) are also required to initiate and sustain DNA synthesis. NTP's are used in the synthesis of RNA primers and ATP is used as an energy source for some of the enzymes needed to initiate and sustain DNA synthesis at the replication fork.



3. Primer

- The enzyme that synthesizes DNA, DNA polymerase, can only add nucleotides to an already existing strand or primer of DNA or RNA that is base paired with the template.

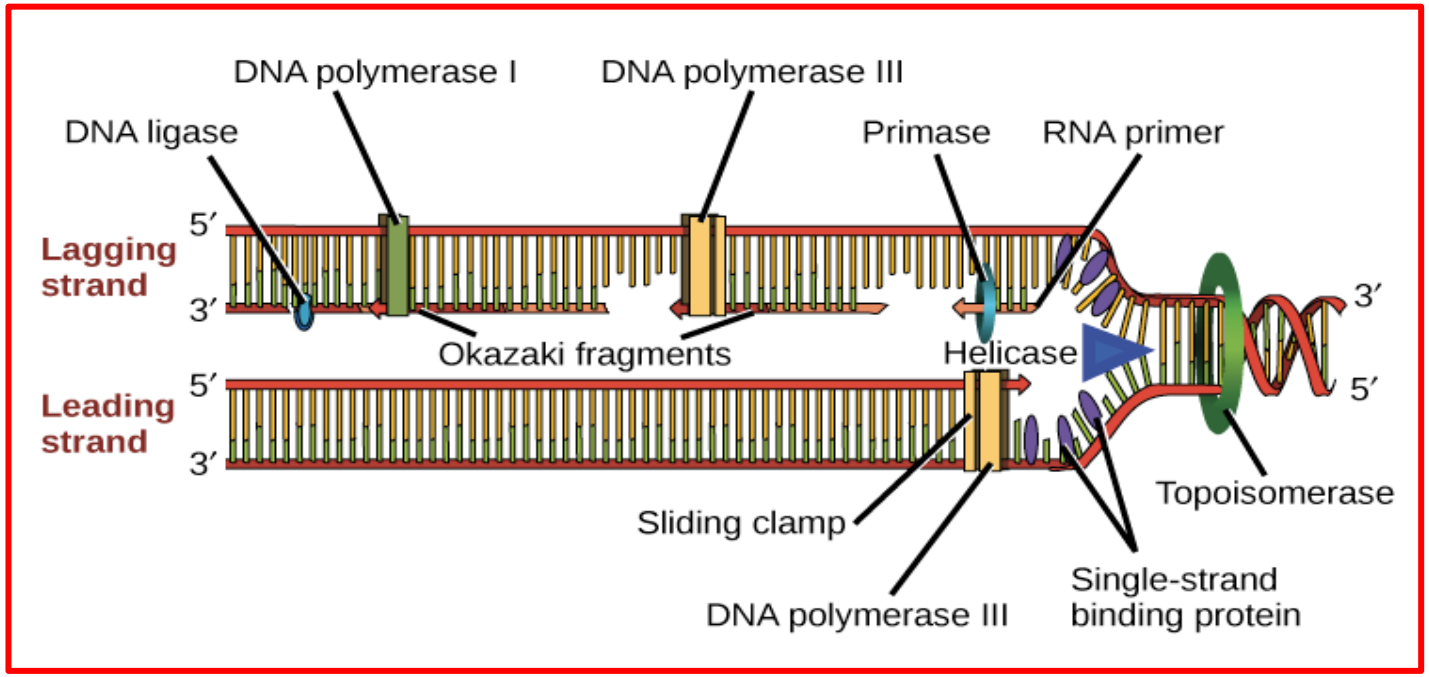




Enzymes

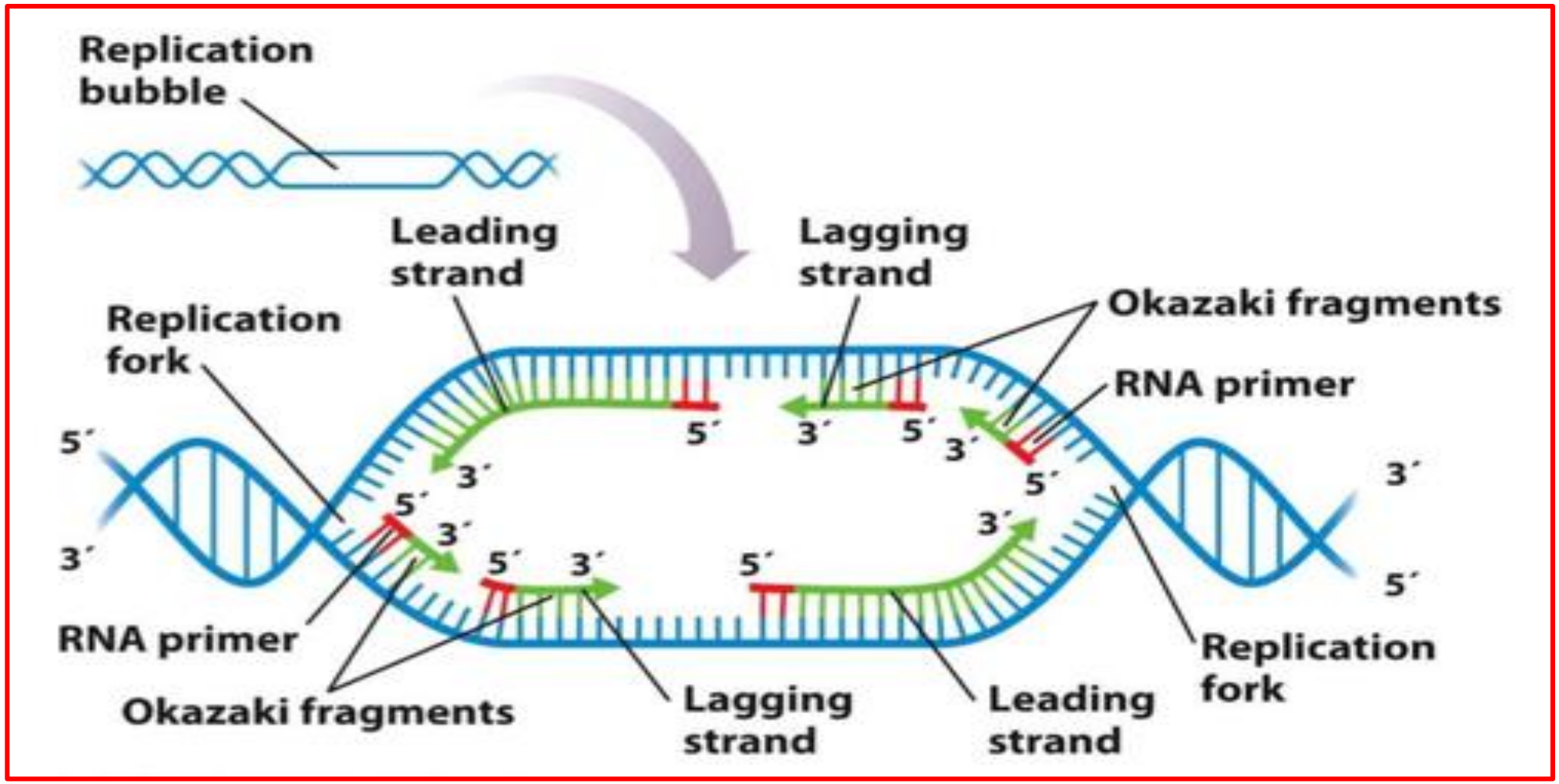
- DNA replication would not occur without enzymes that catalyze various steps in the process. Enzymes that participate in the eukaryotic DNA replication process include:
- **DNA helicase -** unwinds and separates double stranded DNA as it moves along the DNA. It forms the replication fork by breaking hydrogen bonds between nucleotide pairs in DNA.
- **DNA primase -** a type of RNA polymerase that generates RNA primers. Primers are short RNA molecules that act as templates for the starting point of DNA replication.
- **DNA polymerases -** synthesize new DNA molecules by adding nucleotides to leading and lagging DNA strands.

- **Topoisomerase or DNA Gyrase** - unwinds and rewinds DNA strands to prevent the DNA from becoming tangled or supercoiled.
- **Exonucleases -** group of enzymes that remove nucleotide bases from the end of a DNA chain.
- **DNA ligase -** joins DNA fragments together by forming phosphodiester bonds between nucleotides.



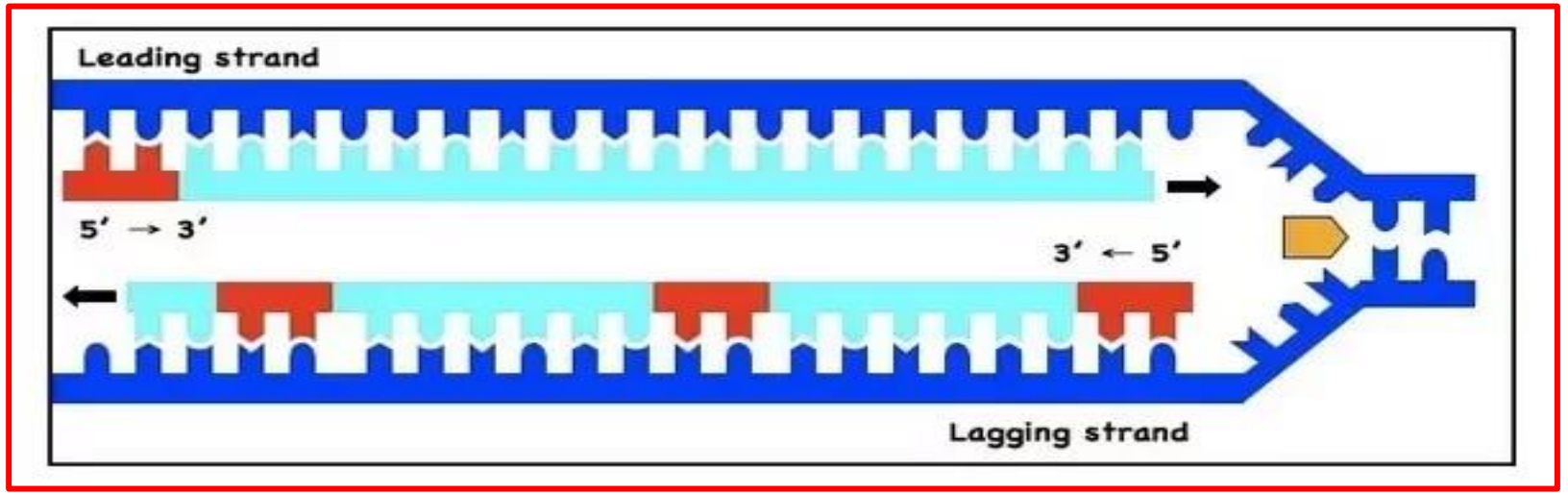
Steps of DNA replication

- **1.** The first step in DNA replication is to ‘unzip’ the double helix structure of the DNA molecule.
- **2.** This is carried out by an enzyme called helicase which breaks the hydrogen bonds holding the complementary bases of DNA together (A with T, C with G).
- **3.** The separation of the two single strands of DNA creates a ‘Y’ shape called a replication ‘fork’. The two separated strands will act as templates for making the new strands of DNA .
- **4.** One of the strands is oriented in the 3’ to 5’ direction (towards the replication fork), this is the leading strand. The other strand is oriented in the 5’ to 3’ direction (away from the replication fork), this is the lagging strand. As a result of their different orientations, the two strands are replicated differently:


Replication bubble
Leading strand
Lagging strand
Okazaki fragments
Replication fork
RNA primer
5´
3´
RNA primer
Okazaki fragments
Lagging strand
Leading strand
Replication fork

Leading Strand

- **5.** A short piece of RNA called a primer (produced by an enzyme called primase) comes along and binds to the end of the leading strand. The primer acts as the starting point for DNA synthesis.
- **6.** DNA polymerase binds to the leading strand and then ‘walks’ along it, adding new complementary nucleotide bases (A, C, G and T) to the strand of DNA in the 5’ to 3’ direction.
- **7.** This sort of replication is called continuous.



Lagging strand

- **5.** Numerous RNA primers are made by the primase enzyme and bind at various points along the lagging strand.
- **6.** Chunks of DNA, called Okazaki fragments, are then added to the
- lagging strand also in the 5’ to 3’ direction.
- **7.** This type of replication is called discontinuous as the Okazaki fragments will need to be joined up later.
- **8.** Once all of the bases are matched up (A with T, C with G), an enzyme called exonuclease strips away the primer(s). The gaps where the primer(s) were are then filled by yet more complementary nucleotides.

- **9.** The new strand is proofread to make sure there are no mistakes in the new DNA sequence.
- 10. Finally, an enzyme called DNA ligase seals up the sequence of DNA into two continuous double strands.
- **11.** The result of DNA replication is two DNA molecules consisting of one new and one old chain of nucleotides. This is why DNA replication is described as semi-conservative, half of the chain is part of the original DNA molecule, half is brand new.
- **12.** Following replication the new DNA automatically winds up into a double helix.