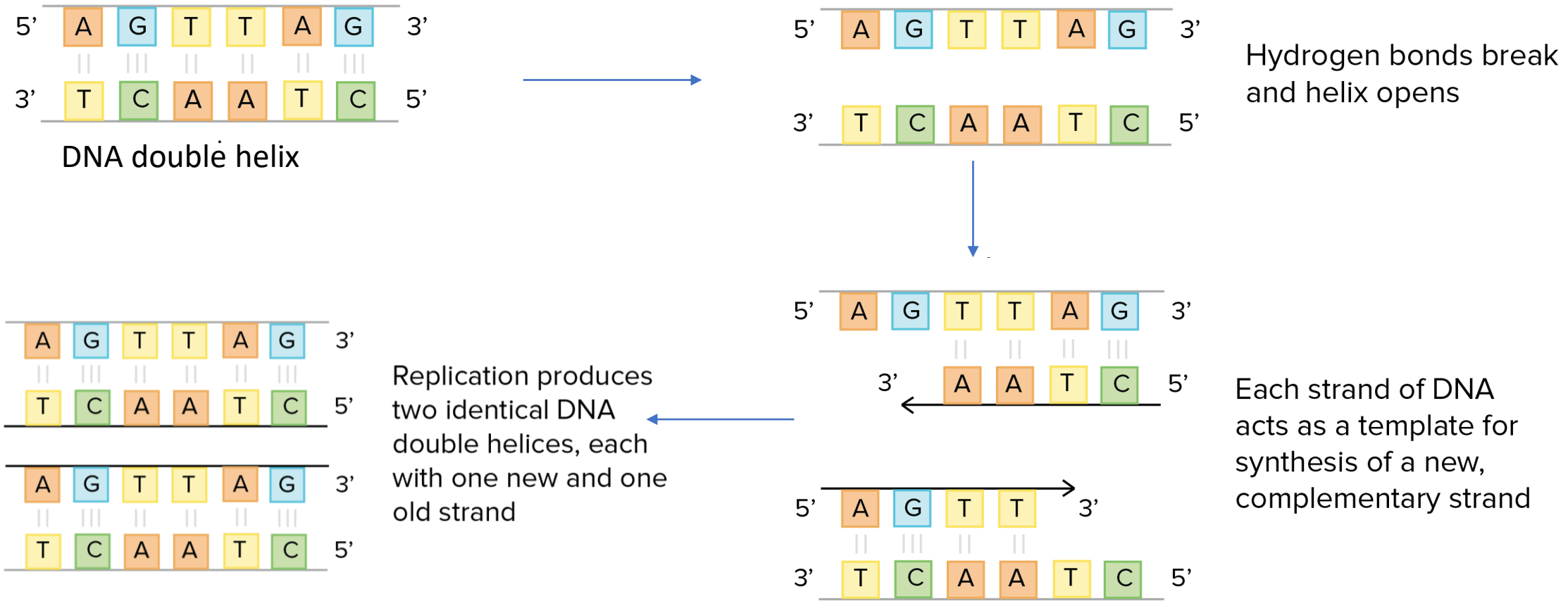
# DNA replication

- DNA replication is **semiconservative**, meaning that each strand in the DNA double helix acts as a template for the synthesis of a new, complementary strand.
- This process takes us from one starting molecule to two "daughter" molecules, with each newly formed double helix containing one new and one old strand.

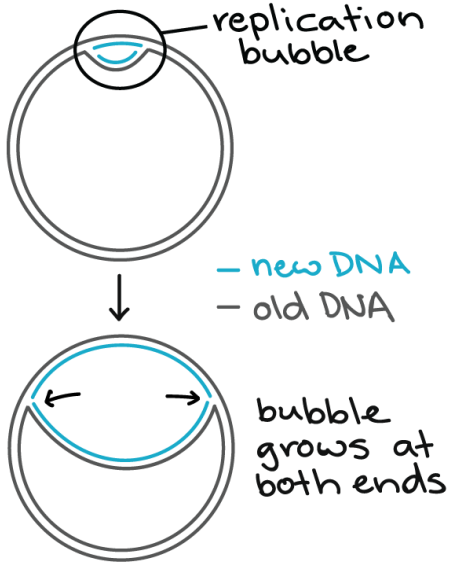


# Origin of replication

- Cells need to copy their DNA very quickly, and with very few errors that occurs with the help of a variety of enzymes and proteins, which work together to make sure DNA replication is performed smoothly and accurately.
- **Starting DNA replication:** Replication always starts at specific locations on the DNA, which are called **origins of replication** and are recognized by their sequence.
- *E. coli*, like most bacteria, has a single origin of replication on its chromosome. The origin is about 245 base pairs long and has mostly A/T base pairs (which are held together by fewer hydrogen bonds than G/C base pairs), making the DNA strands easier to separate.

# Replication fork

- Specialized proteins recognize the origin, bind to this site, and open up the DNA. As the DNA opens, two Y-shaped structures called **replication forks** are formed, together making up what's called a **replication bubble**. The replication forks will move in opposite directions as replication proceeds.



# Helicase and single strand binding proteins

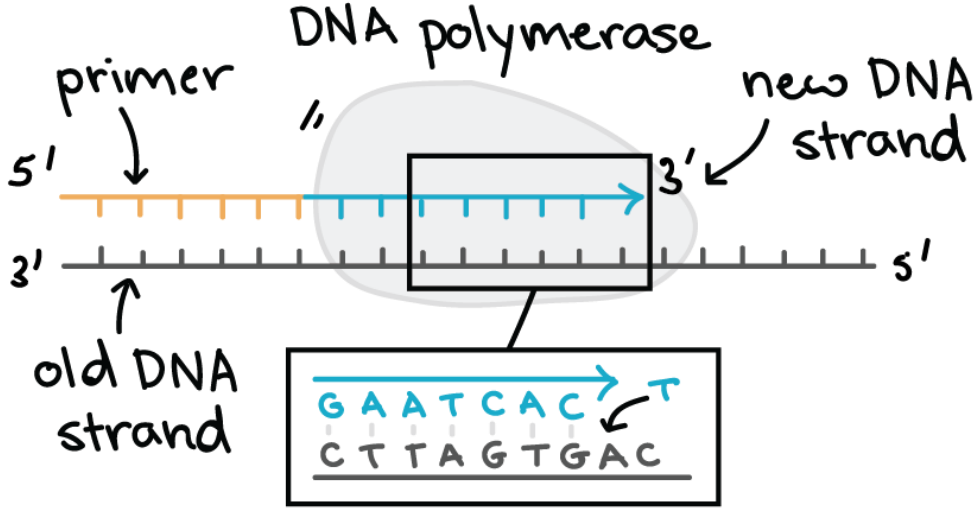
- **Helicase** is the first replication enzyme to load on at the origin of replication. Helicase's job is to move the replication forks forward by "unwinding" the DNA (breaking the hydrogen bonds between the nitrogenous base pairs).
- Proteins called **single-strand binding proteins** coat the separated strands of DNA near the replication fork, keeping them from coming back together into a double helix.

# Primers and DNA polymerase

- **Primers and primase:** Primase makes an RNA **primer**, or short stretch of nucleic acid complementary to the template, that provides a 3' end for DNA polymerase to work on. A typical primer is about five to ten nucleotides long. The primer starts DNA synthesis.
- **DNA polymerase:** DNA polymerases are responsible for synthesizing DNA: they add nucleotides one by one to the growing DNA chain, incorporating only those that are complementary to the template.
- Once the RNA primer is in place, DNA polymerase "extends" it, adding nucleotides one by one to make a new DNA strand that's complementary to the template strand.

# DNA polymerase: key features

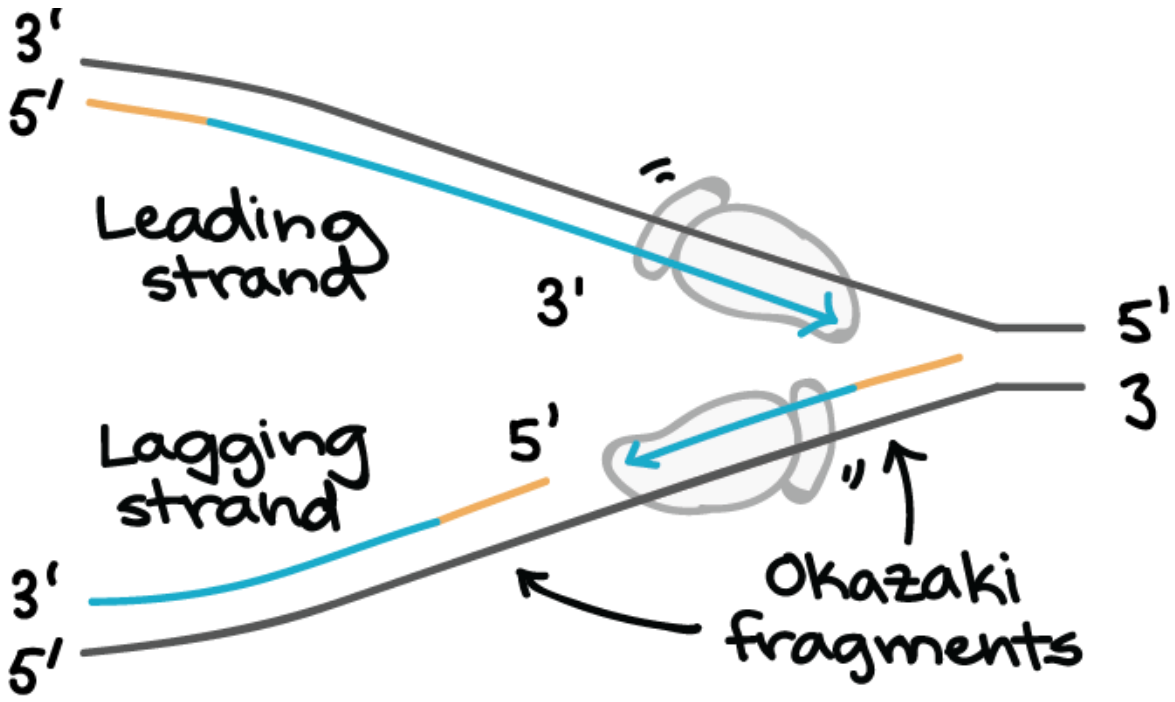
- They always need a template
- They can only add nucleotides to the 3' end of a DNA strand
- They can't start making a DNA chain from scratch, but require a pre-existing chain or short stretch of nucleotides called a **primer**
- They **proofread**, or check their work, removing the vast majority of "wrong" nucleotides that are accidentally added to the chain
- The addition of nucleotides requires energy. This energy comes from the nucleotides themselves, which have three phosphates attached to them (much like the energy-carrying molecule ATP). When the bond between phosphates is broken, the energy released is used to form a bond between the incoming nucleotide and the growing chain.



# Leading and lagging strands

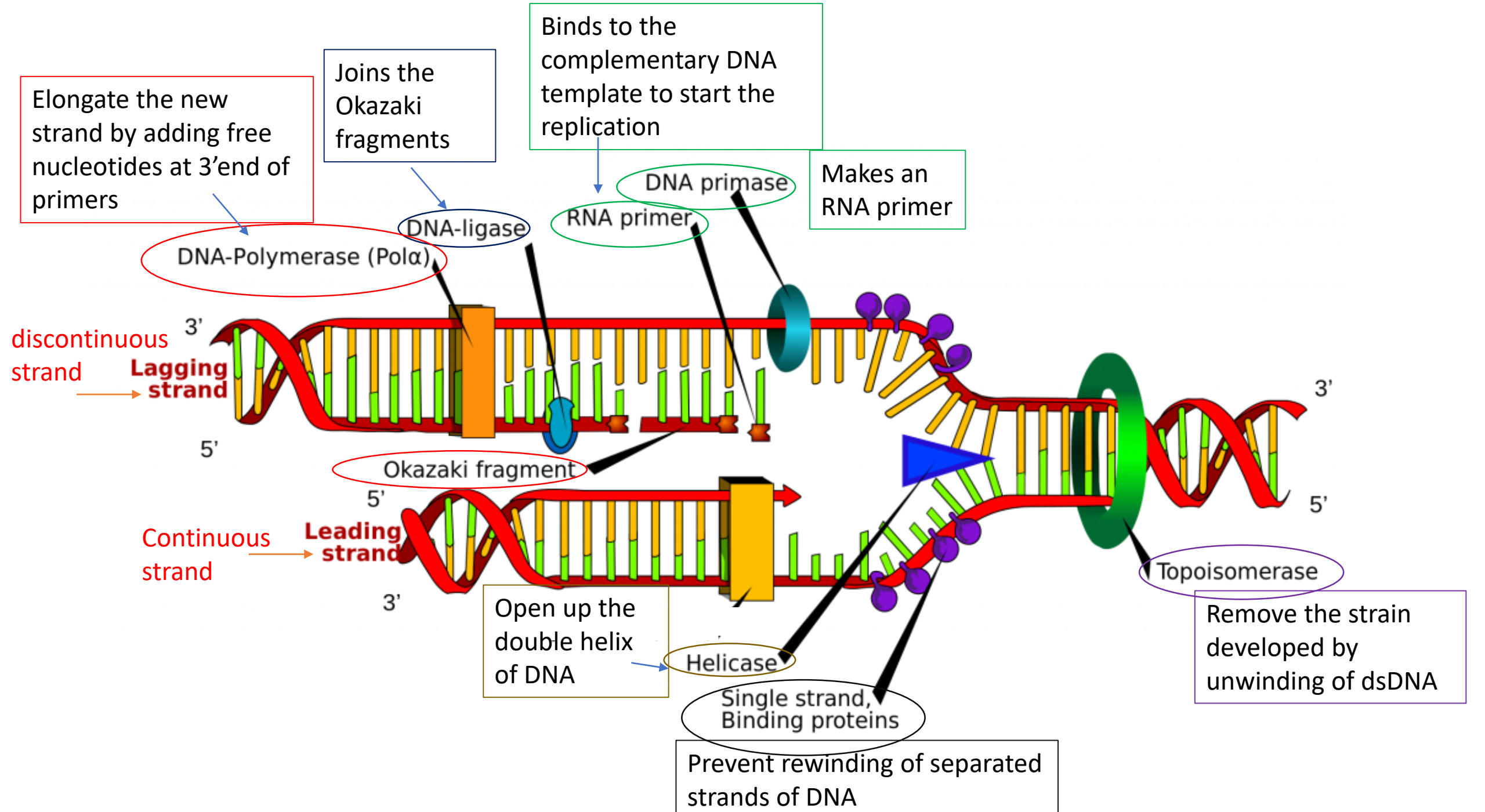
- DNA polymerases can only make DNA in the 5' to 3' direction
- A DNA double helix is always anti-parallel; in other words, one strand runs in the 5' to 3' direction, while the other runs in the 3' to 5' direction
- One new strand, which runs 5' to 3' towards the replication fork is made continuously, because the DNA polymerase is moving in the same direction as the replication fork. This is the **leading strand**.
- The other new strand, which runs 5' to 3' away from the fork is made in fragments, called the **lagging strand**.
- The small fragments are called **Okazaki fragments**, named for the Japanese scientist who discovered them. The leading strand can be extended from one primer alone, whereas the lagging strand needs a new primer for each of the short Okazaki fragments.

# Leading and lagging strand



# Important points

- DNA replication is **semiconservative**. Each strand in the double helix acts as a template for synthesis of a new, complementary strand.
- New DNA is made by enzymes called **DNA polymerases**, which require a template and a **primer** (starter) and synthesize DNA in the 5' to 3' direction.
- During DNA replication, one new strand (the **leading strand**) is made as a continuous piece. The other (the **lagging strand**) is made in small pieces.
- DNA replication requires other enzymes in addition to DNA polymerase, including **DNA primase**, **DNA helicase**, **DNA ligase**, and **topoisomerase**.


Elongate the new strand by adding free nucleotides at 3'end of primers
Joins the Okazaki fragments
Binds to the complementary DNA template to start the replication
DNA primase
Makes an RNA primer
DNA-ligase
RNA primer
DNA-Polymerase (Polα)
discontinuous strand
Lagging strand
3'
5'
Okazaki fragment
5'
Continuous strand
Leading strand
3'
3'
5'
Topoisomerase
Remove the strain developed by unwinding of dsDNA
Open up the double helix of DNA
Helicase
Single strand, Binding proteins
Prevent rewinding of separated strands of DNA