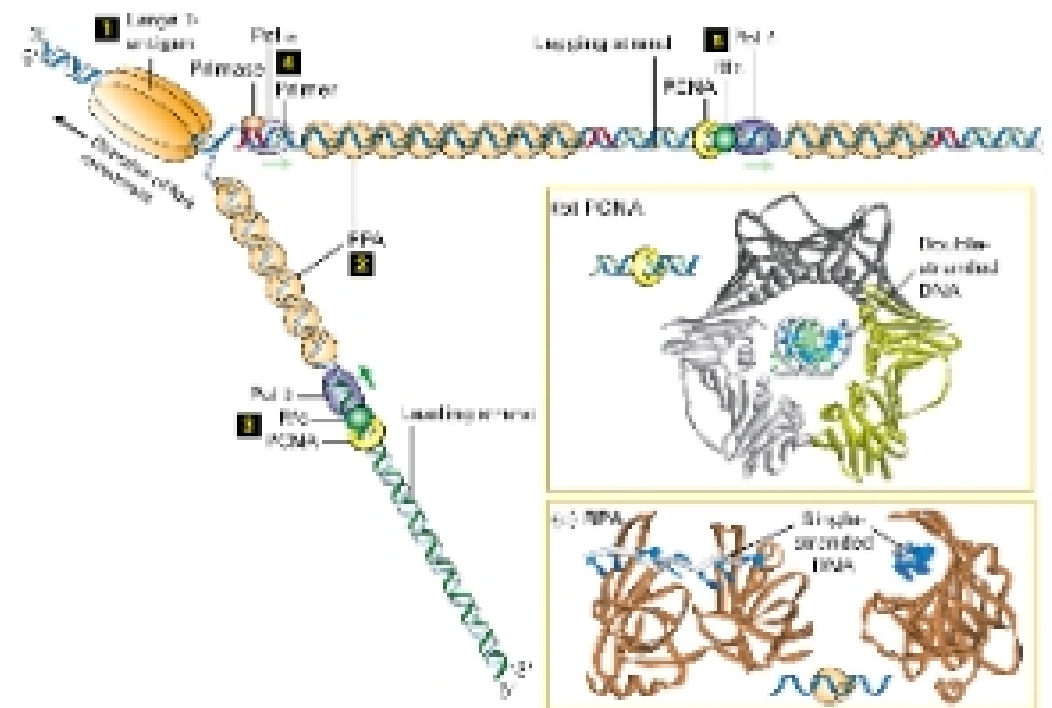


# What is the replication fork? Describe the DNA replication mechanism, starting from the formation of this structure.

- replication fork
    - transition region b/tw template DNA & newly synthesized DNA
    - moves  $5' \rightarrow 3'$
    - usually bi-directional
1. Large T-antigen moves  $5' \rightarrow 3'$  as a DNA helicase, unwinding the DNA & creating the replication fork.
  2. RPA acts as a ss-DNA binding protein on each of the 2 single strands, preventing re-hybridization.
  3. DNA primase & DNA polymerase  $\alpha$  binds each single strand to synthesize RNA primer.
  4. RF-C (clamp loader) displaces DNA polymerase  $\alpha$  (DNA primase) from template.
  5. RF-C recruits PCNA (sliding clamp), which allows DNA polymerase  $\delta$  to be processive & synthesize leading or lagging strand (complementary to single strands produced by replication fork).



## Which strategy is utilized to solve the “over-winding” issue during DNA replication?

- **PROBLEM:** Unwinding of DNA ahead of replication fork produces “torsional tension” (over-winding).
- **SOLUTION:** SWIVEL is a topoisomerase that relieves over-winding in DNA ahead of the replication fork.

# Describe the mechanism of ORC activation in the cell cycle.

1. In G1, Mcm, Cdc6p, & Cdt1p are loaded @ ORI•ORC.
  - Mcm2p-Mcm7p = bind ORI•ORC
  - Mcm4p/Mcm6p/Mcm7p = have helicase activity
  - Cdc6p = must bind chromatin BEFORE Cdt1p is recruited
  - Cdt1p = binds Mcm2p-Mcm7p
2. In S phase, Cdc6p & Cdt1p recruit Mcm (helicase) activator to promote initiation of replication fork.

