DNA Replication in Prokaryotes

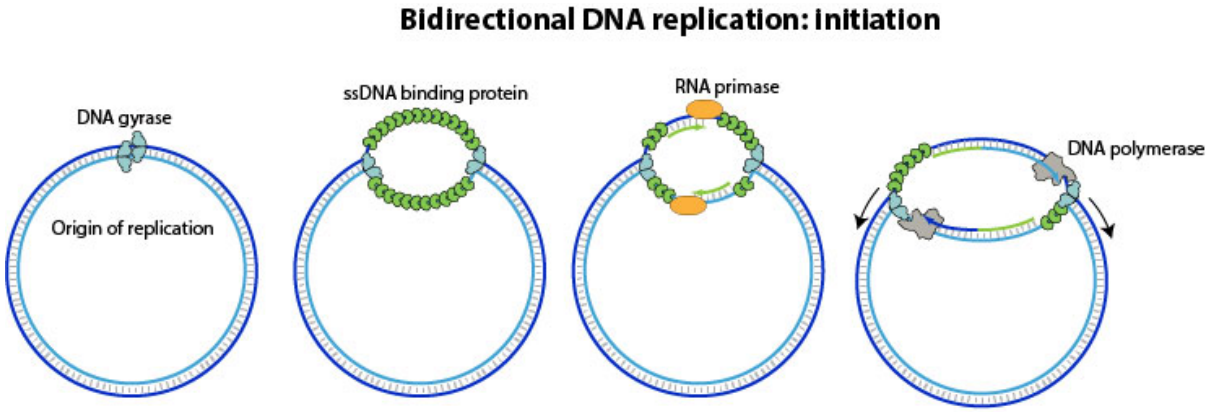
Sharmila Lakshmi T. V.
M.Sc. BIOTECHNOLOGY
Shri Sakthikailassh Women's College, Salem, Tamil Nadu

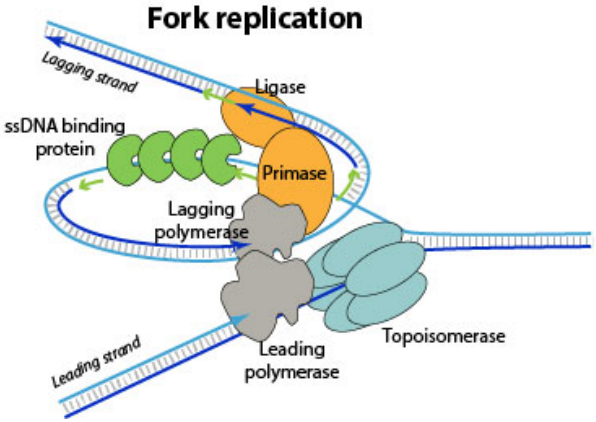
DNA replication

The process by which a double stranded DNA molecule is copied to produce two identical DNA molecule.

- It is an essential process because whenever a cell divides , two daughter cells must contain the same genetic information or DNA as the parent cell.
- In bacterial chromosomes , the replication begins at a specific site called **origin of replication**
- Prokaryotes has only **one origin of replication** site
- At origin site, they have sequence where many proteins bind to initiate the replication (Ori C)
- After binding, they starts to replicate from the origin outward in opposite direction- **bidirectional**

BIDIRECTIONAL REPLICATION



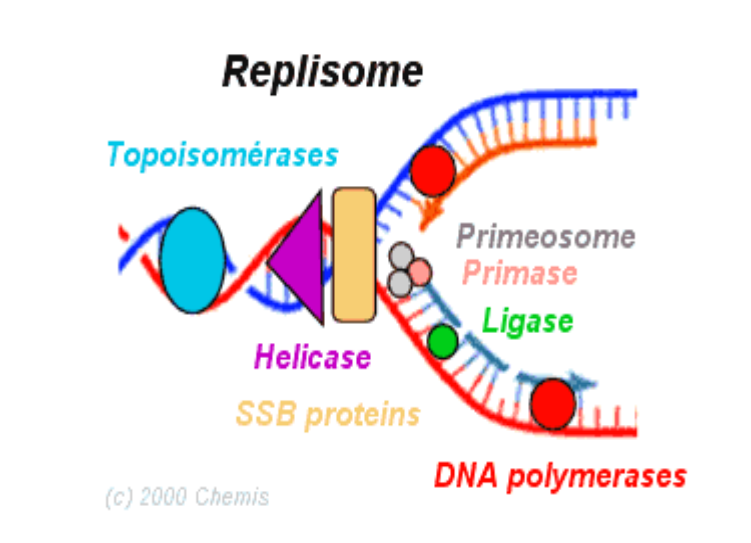


REPLICATION FORK

- The site in which the newly synthesized duplex come together and join the non replicated DNA called replication fork
- It has 2 sites

1. Unwinding of parental double helix duplex
2. Incorporation of nucleotide to newly synthesized complementary strand

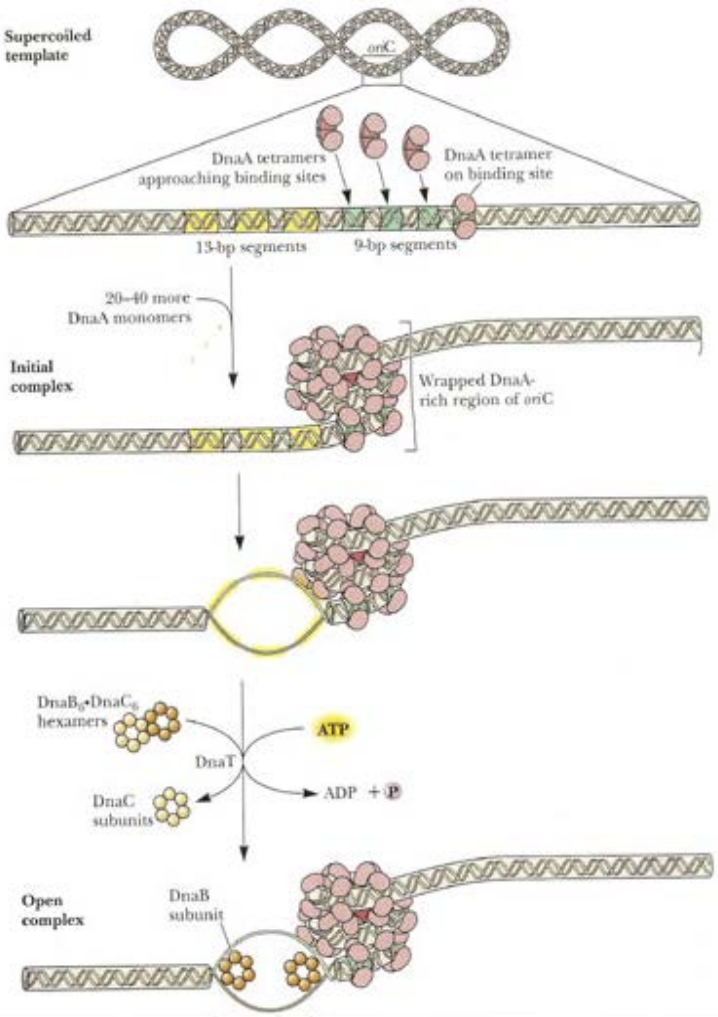
- They replicate in both direction until 2 replication fork meet each other
- When they meet at specific site , they detach from one another and are directed into 2 different cells.

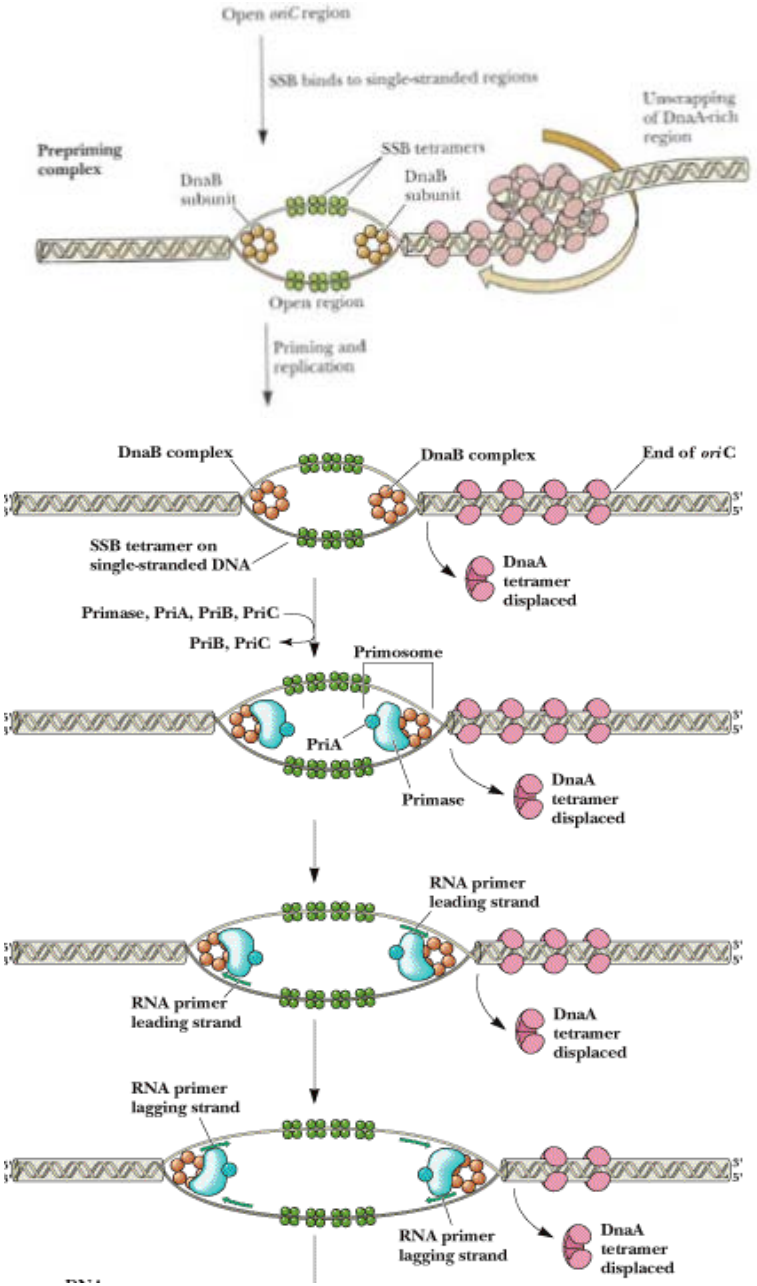


Steps involved in replication

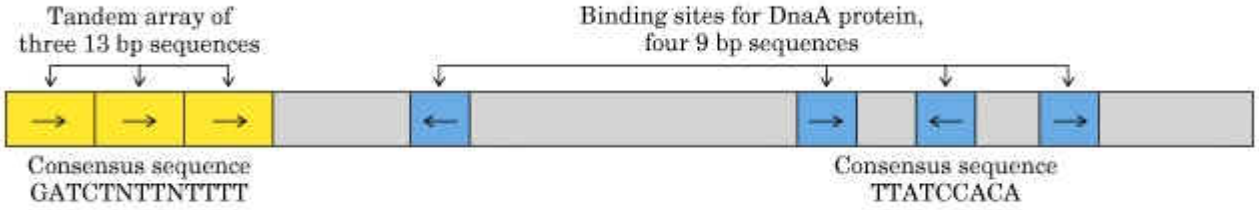
1. Initiation
2. Elongation
3. Termination

INITIATION



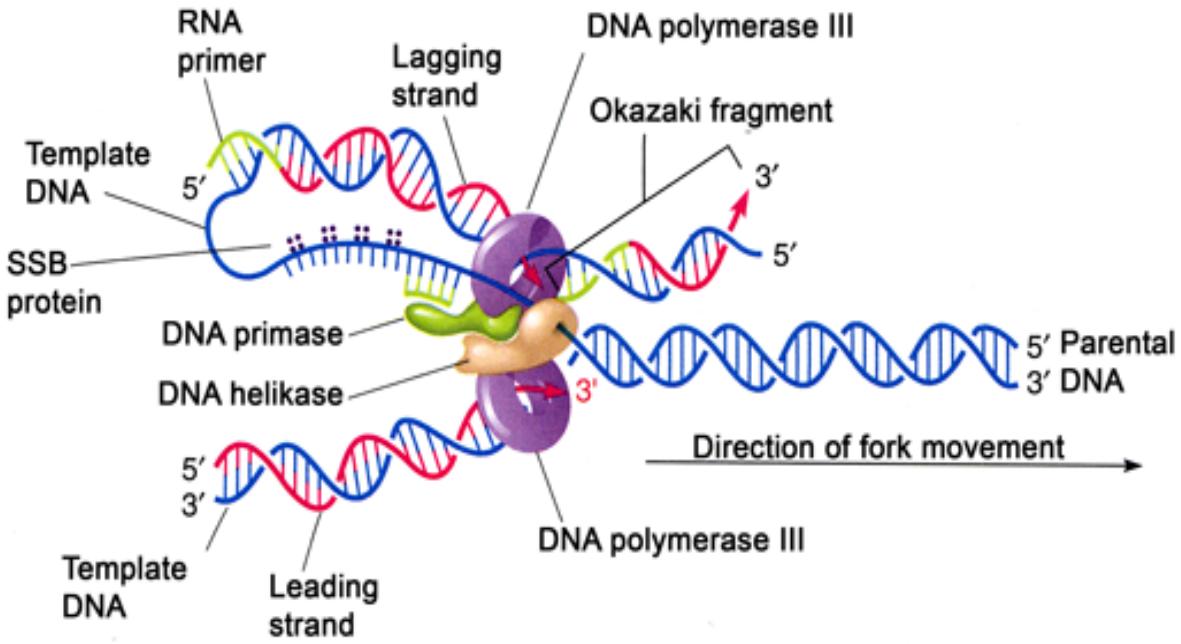


- Replication in E.coli requires 6 proteins – DNA A ,B,C, Helicase, gyrase and SSB
- Initially DNA A molecules recognize the **Ori C** and 2-4 molecules will bind



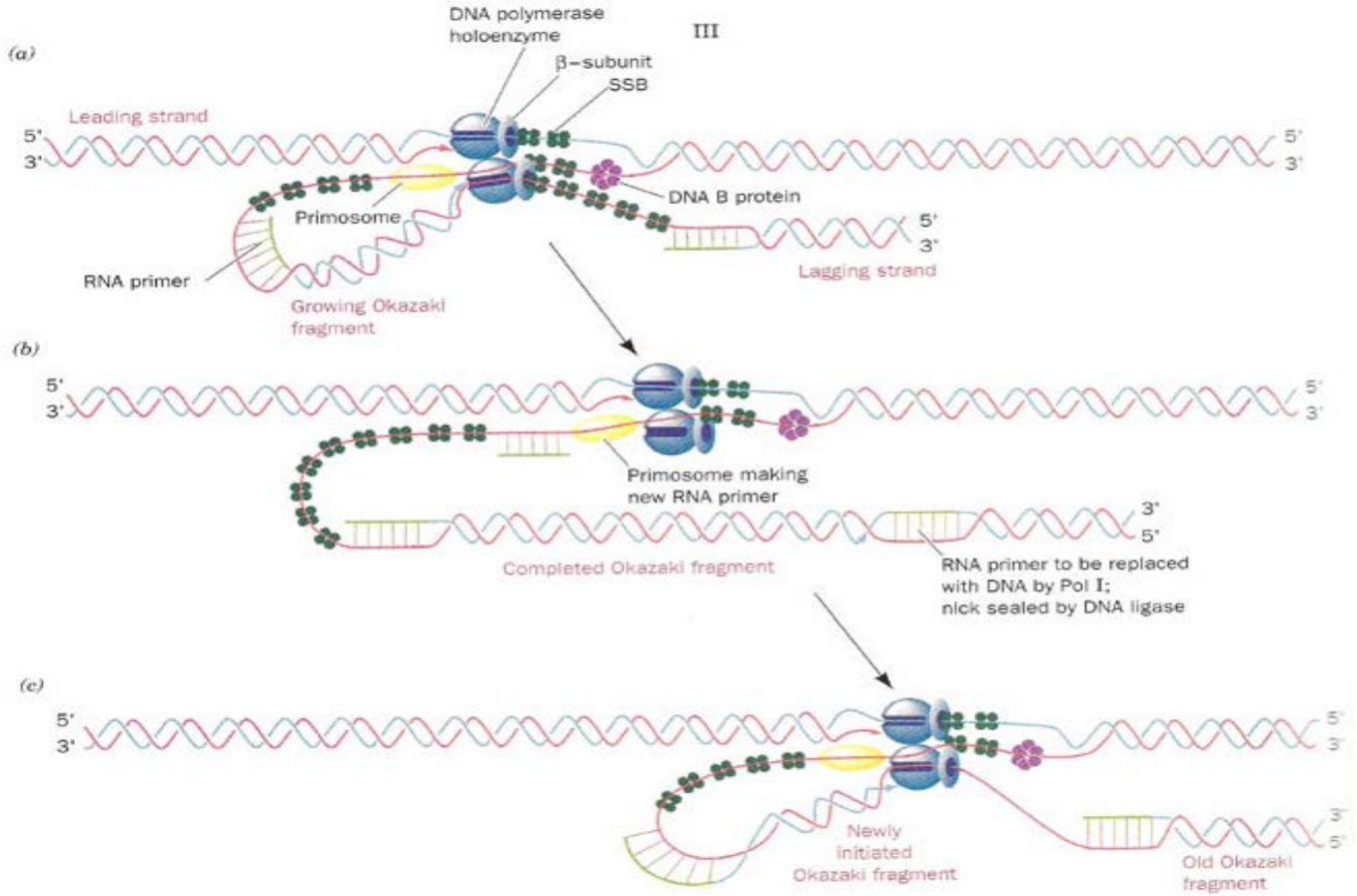
- This facilitates the binding of DNA B-C complex and unwinds the DNA
- **DNA B** serves as **DNA helicase**
- DNA unwinding by helicases are aided by **SSB** (single stranded binding protein)and unwinds the ds DNA
- This facilitated by **DNA gyrase**
- SSB helps in preventing rewound of unwind strand
- At replication fork, **primer** synthesis is initiated by **primase** at origin of replication
- In lagging strand, it occurs at intervals of 1000-2000 bases

- Helicase and primase forms a complex called **primosome**
- Short RNA primer begins to form new **okazaki** fragment in lagging strand
- This results in binding of DNA polymerase III and activates the synthesis of new complementary strand



INITIATION COMPLEX

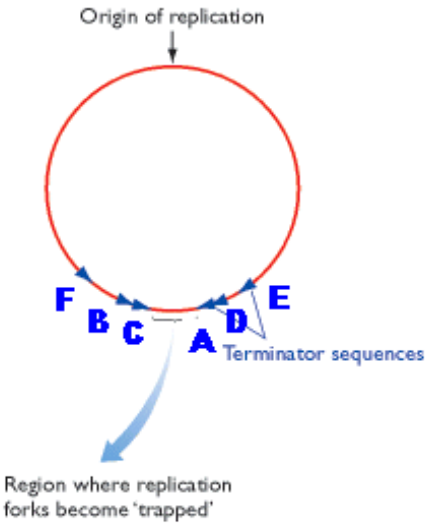
ELONGATION



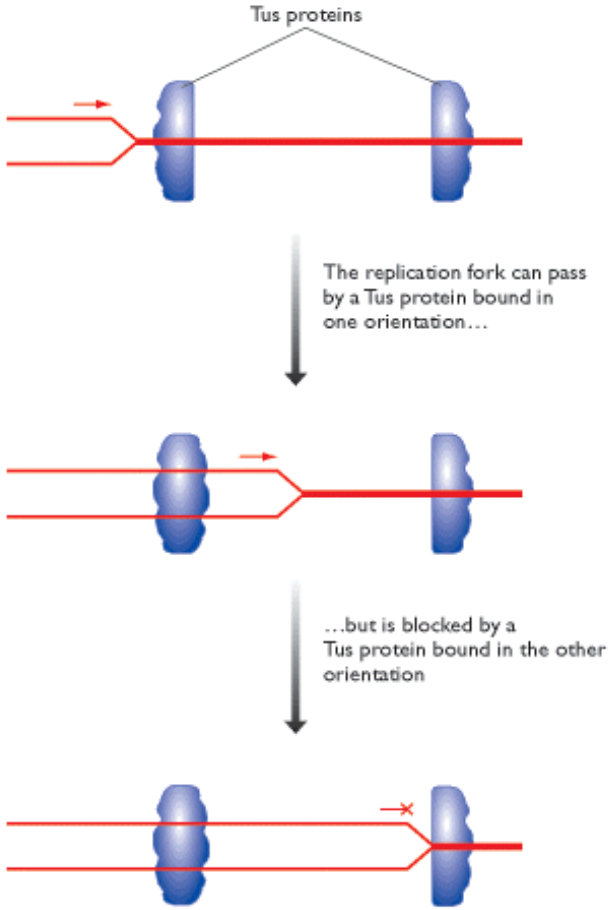
- DNA polymerase III is a holoenzyme contain β sliding clamp
- β clamp helps in association of polymerase with template DNA
- In leading strand, the β sliding clamp attached to DNA and moves by adding one nucleotide to next without diffusing away from template
- In lagging strand , template completes the synthesis of okazaki fragment
- Polymerase III disengages from β clamp and cycled to the new one
- DNA polymerase I involved in DNA repair mechanism
- It has both exonuclease and endonuclease activity
- **Okazaki fragments** will be joined by **DNA LIGASE**

TERMINATION

(A) Terminator sequences in the *E. coli* genome



(B) The role of Tus



- Termination is signaled by specific sequence called **Ter elements**
- They helps in binding termination protein called **Tus protein**
- Tus protein stops DNA B from unwinding of DNA
- Replicated duplexes detaches from one another

References

- https://www.nature.com/scitable/definition/replication-33
- https://www.ncbi.nlm.nih.gov/books/NBK26826
- https://en.wikipedia.org/wiki/Origin_of_replication
- https://www.ncbi.nlm.nih.gov/books/NBK26826
- https://en.wikipedia.org/wiki/Okazaki_fragments
- oregonstate.edu/instruction/bb331/lecture07/lecture07.html
- https://www.ebi.ac.uk/interpro/entry/IPR001001
- https://en.wikipedia.org/wiki/DNA_clamp

THANK YOU