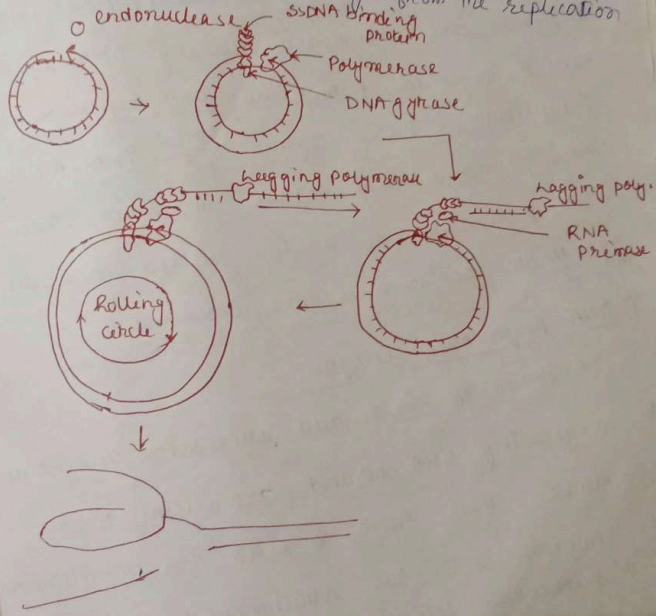
synthesis by primase.

4. The lagging strand RNA primer are removed and okazaki fragment ligated.

5. The replication fork go on until they reach the end of linear genome, or until they meet at the opposite side of a circular genome.

6. After synthesis, topoisomerase allows separation of two strands resulting from the replication



Enzymology of DNA replication / Enzyme associated with DNA replication / Kornberg hypothesis on enzymology :

Enzyme involved in DNA replication was first given by Arthur Kornberg and his collegues in 1957. Kornberg and his collegues isolated an enzyme from *E. coli* which prod DNA synthesis, which later be known as DNA polymerase-1 . ≃ 20 amino acid (proteins) are enzymes are required during DNA replicat which together known as DNA replicase syste (replisome). This enzymes are – i) DNA polym -ase, ii) Polynucleotide ligase, iii) Exonuclease, while a number of protein called helicase, gyrase, or topoisomerase are involved.

i) DNA polymerase enzyme:

Enzymes ~~th~~ that add successive nucleotides to a growing DNA strand are called DNA polymerase. These have 2 sites for the attachm

1. Template site for attachment template DNA
2. 5' triphosphate site for " of deoxyribonucle-

side 5' triphosphate

3. Primer terminus site (For attachment 3'OH end of the DNA primer).

DNA polymerase associated with dNTP nucleotide to primer DNA in direction from 5'- PO4 end to the 3'-OH end. There are 3 DNA polymerase enzyme participate ——

A. DNA Polym. 1 (Pol.1)

B. DNA polymerase - 11 (Pol.11)

C. DNA polymerase -111 (Pol.111)

A. DNA polymerase 1 : The enzyme is spherical with a diameter of about 6.5 nm. It has molecular weight of 1,90,000 and is formed of single polypeptide chain. It has sulphydroxyl group, single inter-chain disulphide and one zinc molecule at active site. DNA pol.-1 molecules is a complex structure formed of —

a) DNA polymerase 3'— 5' exonuclease

b) 5'— 3' exonuclease.

-There are five binding sites of

A. DNA poly.1 :

→ Template site for binding the template DNA.

→ Primer site for binding primer strand of DNA.

→ Primer terminus site for 3'-hydroxyl terminus of primer.

→ ~~Pri~~ 5'-triphosphate site, locus for incoming DNA 5'- " group.

→ 5'→3' exonuclease site, a locus for 5'→3' exonuclease activity situated in the path of growing chain.

Function:

1. 5'→3' polymerase activity: It catalyse the addition of mononucleotide units (deoxyribonucleotide residue) to the free 3'hydroxyl end of polynucleotide chain of DNA for the growth of polynucleotide chain in 5'→3' direction. A pure DNA pol.-1 can add about

1000 nucleotide residue per minute per molecule of at 37°C.

2. 5′ → 3′ exonuclease activity: It catalyse 5′ → 3′ exo-nuclease activity in DNA repair by removing and replacing damaged base pair. It also catalyses remo-val of pieces of RNA primers used in DNA rep-lication from the 5′ end of each okazaki fragment

3. 3′ → 5′ proof reading exonuclease activity: This activity of polymerase-I has a proof reading correc-ting function. It removes the incorrect nucleotide nucleotide and places the correct one

B. DNA pol. II:

It is comparatively small enzyme with a molecu-lar weight of about 1,20,000 daltons. Its biological function is not fully established. It seems to repair DNA by filling polynucleotide chain. Thus the pol.-II can't replicate long strand but can fill gaps in DNA duplex

C. DNA pol. III :

This enzyme was discovered by T. Kornberg and M.L. Gefter in 1972. It is largest, most complex and most active polymerase enzyme out of three. Molecular weight = 9,00,000 and consist of 18-20 subunits. Because it role as primary role in replication, so it is refers as replicase.