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FACULTY OF ENGINEERING & TECHNOLOGY
DEPARTMENT OF BIOTECHNOLOGY

Basic Requirements of DNA Replication

- The basic requirements and components of replication are the same for prokaryotes and eukaryotes.
- Replication in prokaryotes is much better understood than in eukaryotes.
- **Substrates:** 4 deoxyribonucleosides triphosphate(dATP , dGTP ,dCTP, dTTP)
- **Template :** separated strands of DNA serve as template for the synthesis of the new daughter DNA strands .It is required to direct the addition of the appropriate complementary nucleotide to newly synthesized DNA strand.
- **Proteins and Enzymes :** different types of DNA polymerases and proteins

Proteins involved in initiation of DNA replication at E. coli origin

Protein	Molecular weight	Number of subunits	Function
DNA A Protein	52000	1	Recognizes ori sequences, opens DNA duplex at a specific site in origin
DNA B Protein (Helicase)	300000	6	Unwinds DNA, primosome constituent
DNA C Protein	29000	1	Required for DNA B binding at origin
HU (Histone like protein)	19000	2	DNA binding protein, stimulates initiation
Primase (DNA G Protein)	60000	1	Synthesizes RNA primers, primosome constituent
SSB (single stranded binding protein)	75600	4	Binds single stranded DNA and stabilizes these separated DNA and prevents renaturation of DNA
DNA topoisomerase II (DNA Gyrase)	400000	4	releases torsional strain generated by DNA unwinding
DNA polymerase I	103000	1	Filling gaps and excisions of primers
DNA polymerase III	791500	17	new DNA strand elongation
DNA ligase	74000	1	Seals the single strand nick between the nascent chain and Okazaki fragments on lagging strand (ligation)

Proteins involved in initiation of DNA replication at E.Coli origin

Protein	Molecular weight	Number of subunits	Function
RNA polymerase	454000	5	Facilitates DNA A Protein binding activity
DNA methylase	32000	1	Methylate of (5') GATC sequence at ori C
DNA polymerase I			Filling gaps and excisions of primers
Ter binding protein			Prevents DNA B Protein(Helicase) from further unwinding of DNA and facilitates the termination of replication

Comparison of Prokaryotic and Eukaryotic DNA polymerases

Prokaryotic DNA polymerases	Eukaryotic DNA polymerases	Functions
I (Pol)	α	Gap filling and synthesis between Okazaki fragments of lagging strand(5' \rightarrow 3' polymerization activity)
II(Poll)	ϵ	DNA proof reading and DNA repair(3' \rightarrow 5' exonuclease activity)
	β	DNA repair (5' \rightarrow 3' exonuclease activity)
	γ	Mitochondrial DNA synthesis
III(Poll)	δ	Functions at replication fork, catalyzing Leading and lagging strand synthesis

