

# Extrachromosomal Replication and types of Recombination



## EXTRACHROMOSOMAL REPLICATION

Plasmids are known as extrachromosomal DNA, and they have self-replicating properties. Plasmids are found in bacteria (Gram-negative and positive both), yeast and some other fungi and are present as covalently closed, circular double-strand DNA. In some cases, the linear plasmid is also reported. Plasmid codes various genetic determinants that help the prokaryote survive in extreme conditions. Some of them also code for some antibiotic resistance genes.

These extrachromosomal DNAs depend on host-encoded molecules specific for their replication. The origin of replication starts from the particular site, known ori site, and replication occurs either by the rolling circle method or theta method. Some plasmids require antisense RNA and repeated sequence for replication located close to the ori site, which determines attributes like copy number and incompatibility of the plasmid.

The **rolling circle** mechanism is commonly found in Gram-negative bacterial plasmids and Phage  $\phi$ \*174. Initiation starts with one strand of double strand circular DNA, and Nicks is created after from 5' end, and DNA starts growing. When it reaches the proper length, it gets displaced, and configuration changes occur, forming newly synthesised circular DNAs.

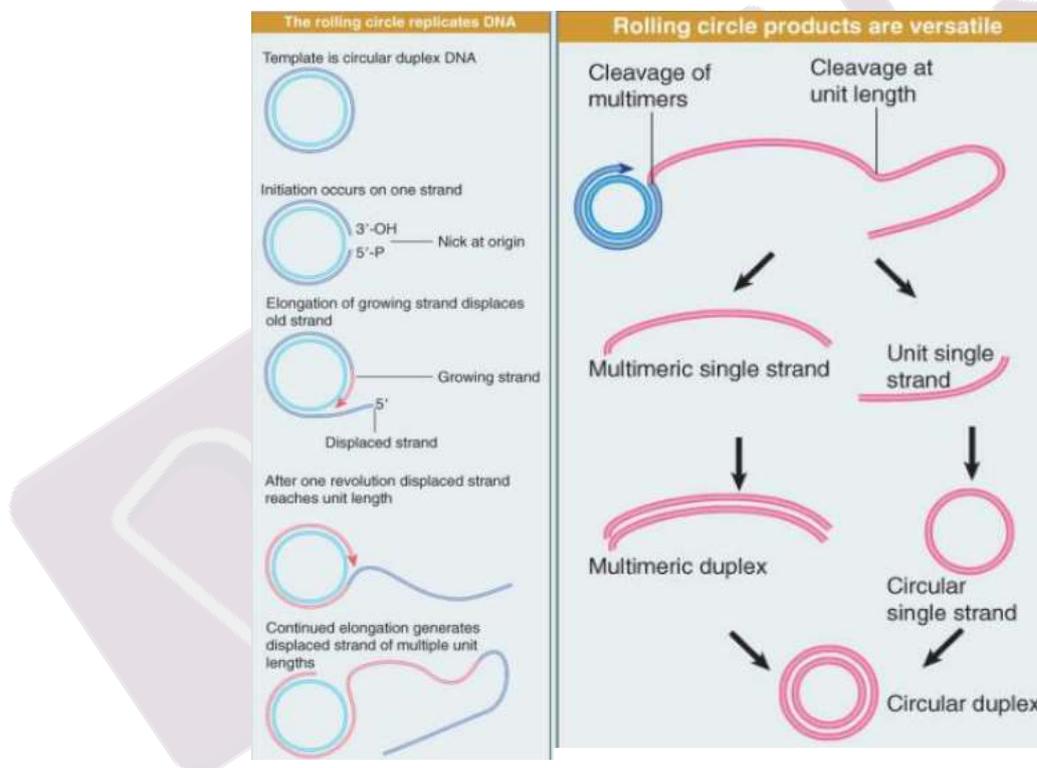


Figure 1. Rolling Circle Model of Replication

## RECOMBINATION

Recombination is when the breakage and joining between two chromosomal fragments occur through which a new combination of genetic material is developed. It leads to the development of variations in population.

## Homologous Recombination

Homologous recombination is a type that includes the exchange of nucleotide sequences between two homologous chromosomes. It takes part in the DNA double-strand break mechanism, and due to this type of mechanism, a new gene combination is formed, which becomes the cause of genetic variation in the population. Horizontal gene transfer is also a reason for causing homologous recombination in which the exchange of genetic material between the strain of bacteria and virus occurs.

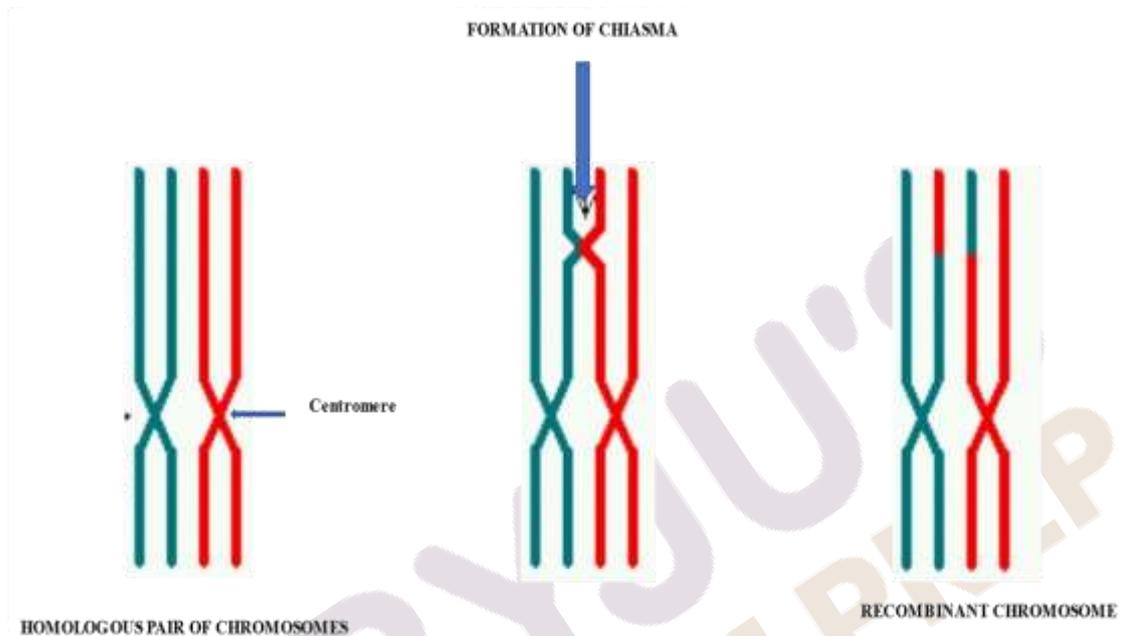


Figure 2. Homologous recombination

Homologous recombination occurs in cells during meiosis, in which the exchange of genes between maternal chromatid and paternal chromatid takes place between homologous chromosome pairs. For this type of recombination to occur, a cross-type structure is formed, which is known as the *Holliday junction*, the name given in honour of Robin Holliday; this model is also known as the heteroduplex model. In this mechanism, a single-strand nick is introduced at the exact position of both parental DNA molecules. Then nicked part is exchanged by complementary base pairing, and as the crossed strand is formed, which is known as the Holliday junction, these hetero duplex results in the exchange of polynucleotide in both strands. After exchanging the nucleotide, the Holliday junction is resolved into its double-strand form by cleavage. This type of recombination is seen in crossing over. Proteins involved in homologous recombination in *Escherichia coli*:

- Rec B- Function as 3'-5' helicase and nuclease
- Rec C- Recognition of Chi site
- Rec D -5'-3; helicase activity

The figure below reveals a double-strand break and synthesis model in meiotic recombination. In this type of mechanism in step 1, double-strand breaks are generated. After this processing, some enzyme complex forms a nucleoprotein filament capable of searching for homology in DNA strand invasion. Afterwards, Rad protein-mediated recombination occurs, which forms a DNA loop (D Loop), allows DNA polymerase activity, and forms Heteroduplex. The nick created in a single invasion may enter into a double-strand break pathway and form a Holliday junction, which can resolve into a crossover product (CO) or a non-crossover (NCO) product. CO product requires the symmetric cleavage of the Holliday junction, and the enzyme resolvase helps. Additionally, NCO products may be resolved by resolvase or other protein. A complex version of this mechanism involves dHj formation, which can resolve Holliday junction as CO and NCO products.

### Site-Specific Recombination

Such a recombination process occurs between the chromosomes that have a less homologous sequence. In this mechanism, short DNA sequences are first recognised after binding, integrating, and excision between

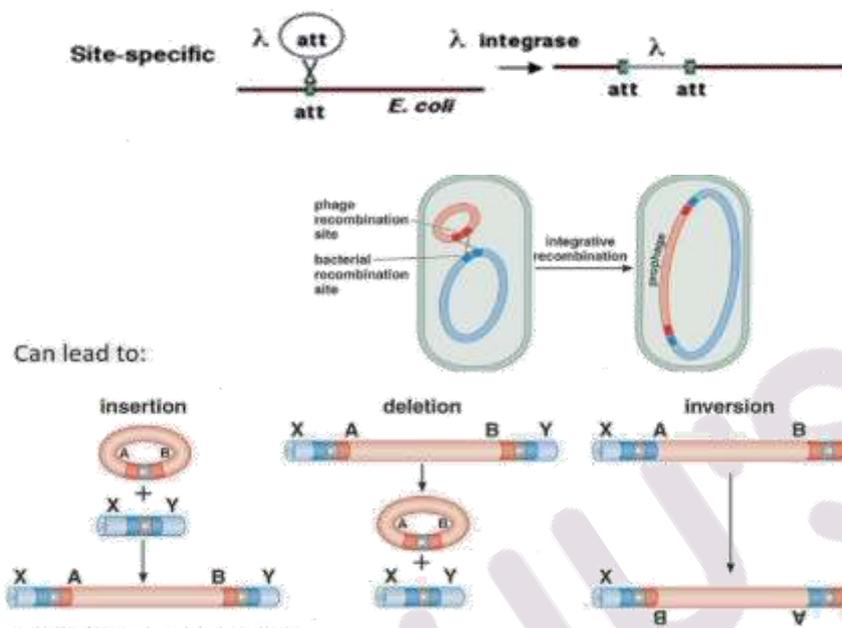


Figure 3. Site specific recombination

phage and bacterial DNA. Site-specific recombination regulates the expression of genes in both prokaryotic and eukaryotic cells. The recombination process initiated by lambda integrase and tyrosine recombinase results in the breakage of the phosphodiester bond, then covalently attaches with the 3' phosphoryl group. In the integration process, IHF Integration of Host factor is required; IHF is a sequence-dependent DNA binding protein that introduces large bend in DNA. IHF binds with the integrase and binding site of the DNA arm. After that, a hybrid version is created at the attachment site known as attL and attR.

Table 1. Differences between the two modes of recombination

	Homologous Recombination	Site-Specific Recombination
<b>Definition</b>	Homologous recombination is genetic recombination in which genetic material is exchanged between two identical molecules of double-stranded or single-stranded nucleic acids (DNA or RNA).	Site-specific recombination is a type of genetic recombination in which DNA strand exchange takes place between DNA segments that possess at least a certain degree of sequence homology but no extensive homology.
<b>Location of recombination</b>	Occurs anywhere within the homology.	Occurs only at specific sites.
<b>Recombination takes place between</b>	Occurs between long DNA strands.	Occurs between short DNA sequences.

<b>Example</b>	General recombination of eukaryotes in meiosis.	Site-specific recombination system that involves when bacteriophage integrates into a bacterial chromosome.  And rearrangement of immunoglobulin genes invertebrate animals.
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