VIVEKANANDA COLLEGE

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NAAC ACCREDITED 'A' GRADE

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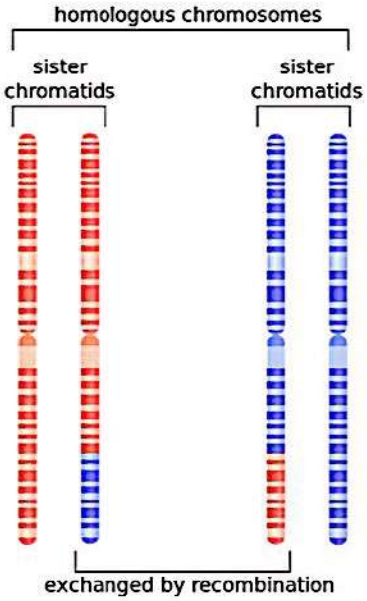
RECOMBINATION OF DNA



Homologous recombination is a type of genetic recombination in which nucleotide sequences are exchanged between two similar or identical molecules of double-stranded or single-stranded nucleic acids (usually DNA as in cellular organisms but may be also RNA in viruses). It is most widely used by cells to accurately repair harmful breaks that occur on both strands of DNA, known as double-strand breaks (DSB). Homologous recombination also produces new combinations of DNA sequences during meiosis, the process by which eukaryotes make gamete cells, like sperm and egg cells in animals. These new combinations of DNA represent genetic variation in offspring, which in turn enables populations to adapt during the course of evolution. Homologous recombination is also used in horizontal gene transfer to exchange genetic material between different strains and species of bacteria and viruses.

Although homologous recombination varies widely among different organisms and cell types, for double-stranded DNA (dsDNA) most forms involve the same basic steps. After a double-strand break occurs, sections of DNA around the 5' ends of the break are cut away in a process called resection. In the strand invasion step that follows, an overhanging 3' end of the broken DNA molecule then "invades" a similar or identical DNA molecule that is not broken. After strand invasion, the further sequence of events may follow either of two main pathways discussed below (see Models); the DSBR (double-strand break repair) pathway or the SDSA (synthesis-dependent strand annealing) pathway. Homologous recombination that occurs during DNA repair tends to result in non-crossover products, in effect restoring the damaged DNA molecule as it existed before the double-strand break.

Homologous recombination is conserved across all three domains of life as well as DNA and RNA viruses, suggesting that it is a nearly universal biological mechanism. The discovery of genes for homologous recombination in protists—a diverse group of eukaryotic microorganisms—has been interpreted as evidence that meiosis emerged early in the evolution of eukaryotes. Since their dysfunction has been strongly associated with increased susceptibility to several types of cancer, the proteins that facilitate homologous recombination are topics of active research. Homologous

recombination is also used in gene targeting, a technique for introducing genetic changes into target organisms.

IN EUKARYOTES

Homologous recombination (HR) is essential to cell division in eukaryotes like plants, animals, fungi and protists. In cells that divide through mitosis, homologous recombination repairs double-strand breaks in DNA caused by ionizing radiation or DNA-damaging chemicals.Left unrepaired, these double-strand breaks can cause large-scale rearrangement of chromosomes in somatic cells, which can in turn lead to cancer.

In addition to repairing DNA, homologous recombination also helps produce genetic diversity when cells divide in meiosis to become specialized gamete cells—sperm or egg cells in animals, pollen or ovules in plants, and spores in fungi. It does so by facilitating chromosomal crossover, in which regions of similar but not identical DNA are exchanged between homologous chromosomes. This creates new, possibly beneficial combinations of genes, which can give offspring an evolutionary advantage.Chromosomal crossover often begins when a protein called Spo11 makes a targeted double-strand break in DNA.These sites are non-randomly located on the chromosomes; usually in intergenic promoter regions and preferentially in GC-rich domains.These double-strand break sites often occur at recombination hotspots, regions in chromosomes that are about 1,000–2,000 base pairs in length and have high rates of recombination. The absence of a recombination hotspot between two genes on the same chromosome often means that those genes will be inherited by future generations in equal proportion. This represents linkage between the two genes greater than would be expected from genes that independently assort during meiosis.

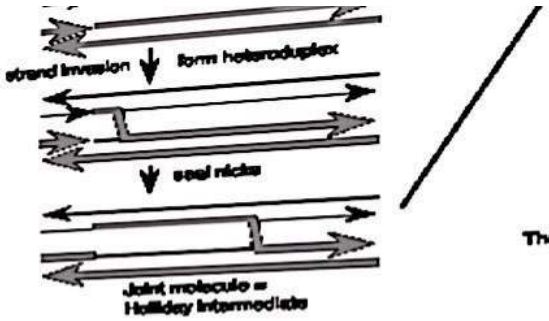
IN BACTERIA

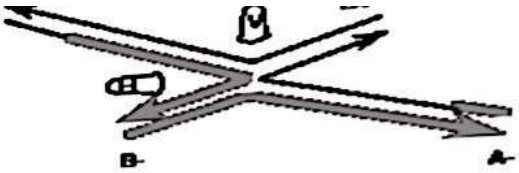
Homologous recombination is a major DNA repair process in bacteria. It is also important for producing genetic diversity in bacterial populations, although the process differs substantially from meiotic recombination, which repairs DNA damages and brings about diversity in eukaryotic genomes. Homologous recombination has been most studied and is best understood for Escherichia coli.Double-strand DNA breaks in bacteria are repaired by the RecBCD pathway of homologous recombination. Breaks that occur on only one of the two DNA strands, known as single-strand gaps, are thought to be repaired by the RecF pathway.Both the RecBCD and RecF pathways include a series of reactions known as branch migration, in which single DNA strands are exchanged between two intercrossed molecules of duplex DNA, and resolution, in which those two intercrossed molecules of DNA are cut apart and restored to their normal double-stranded state.

HOLLIDAY MODEL

In 1964, Robin Holliday proposed a model that accounted for heteroduplex formation and gene conversion during recombination. Although it has been supplanted by the double-strand break model (at least for recombination in yeast and higher organisms), it is a useful place to start. It illustrates the critical steps of pairing of homologous duplexes, formation of a heteroduplex, formation of the recombination joint, branch migration and resolution.

The steps in the Holliday Model are illustrated in the following Figure



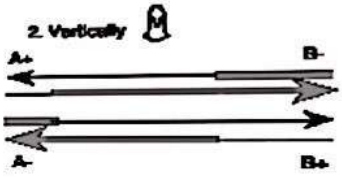


The joint molecule can be resolved in either of 2 ways:

1. Horizontally

OR

2. Vertically



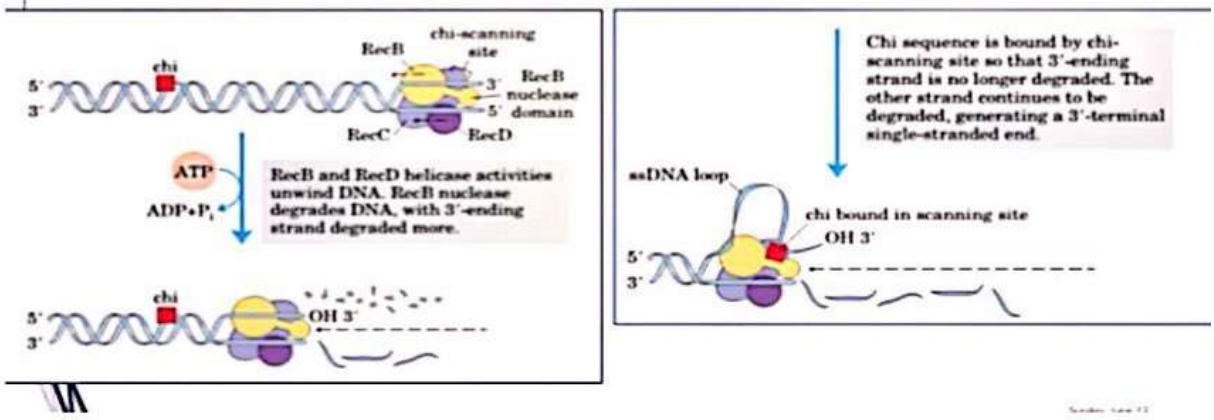
A region of heteroduplex is left, but the

The recombination intermediate is then resolved by nicking a strand in each duplex and ligation.

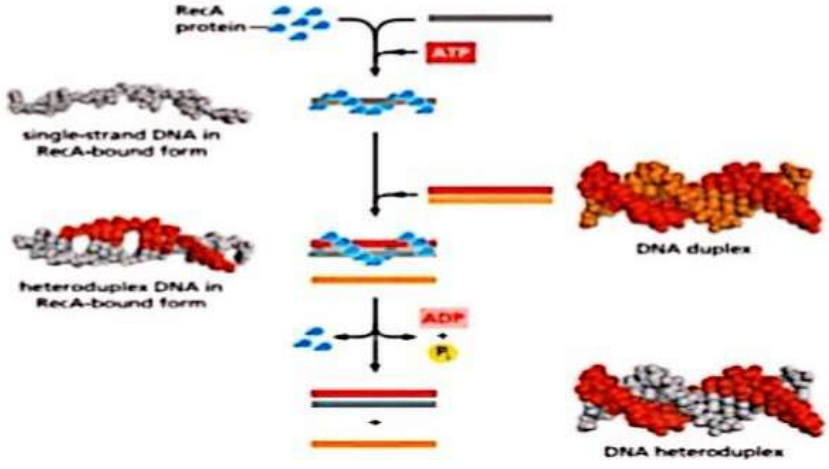
Recombination requires host of enzymes and other proteins

- Ruv A and Ruv B proteins form a complex that binds to Holliday intermediates, displaces RecA protein, and promote branch migration at higher rates that does RecA.
- Nucleases that often cleaves Holliday intermediates, often called resolvases, has been isolated from bacteria and yeast.

Nuclease and helicase activity of RecBCD enzyme



DNA strand invasion catalyzed by RecA protein



diated DNA strand