BICH/GENE 431 KNOWLEDGE OBJECTIVES

Chapter 10 – Homologous Recombination

DNA recombination is process by which segments of DNA are exchanged or moved between or within DNA duplexes.

Two major types of recombination

- homologous recombination
- site-specific recombination involving recombinases or transposons (next chapter)

Holliday model for homologous recombination – understand Fig. 10-2

- alignment of homologous regions
- ds break or nicks
- strand invasion
- formation of Holliday junction
- heteroduplex formation
- branch migration
- resolution to give crossover products or patch recombination (without crossover)

Recombinational repair of dsDNA breaks – know mechanism

E. coli enzymes used in homologous recombination

- rec mutants in E. coli deficient in generalized recombination
- E. coli does not have enzyme to break DNA for recombination – relies on ds breaks already or disassembly of replication fork after encountering a nick
- RecBCD complex – endonuclease and helicases to process ends
- chi sites: how do they work along with RecBCD? polarity (orientation) of these sites
- RecA protein forms filaments on DNA processed by RecBCD
    - central player in homologous recombination
    - cooperative binding in a polar fashion
    - extends DNA after binding
    - catalyzes single-strand uptake reaction: know examples of such reactions analyzed in vitro
    - what does RecA require in order to work?
- RuvAB complex binds Holliday junction and promotes branch migration
  RuvB is helicase
- RuvC is nuclease to resolve Holliday junctions

Meiotic recombination in eukaryotes

- Spo11 protein for breaking DNA
- MRX complex for processing DNA ends
- Rad51 and Dmc1 are RecA homologs
- Plus many other proteins needed too.

Gene conversion results from mismatch repair within heteroduplex formed during meiotic recombination.