PCB 3063 with Dr. Bass- Exam #3 Study Guide (Ch. 8-12)

Chapter 8- Prokaryotic Genetics

- **Bacteriophages**: viruses that use bacteria as their host
- Bacteria and viruses are extremely useful for research because of their short reproductive cycle and are studied in pure cultures.

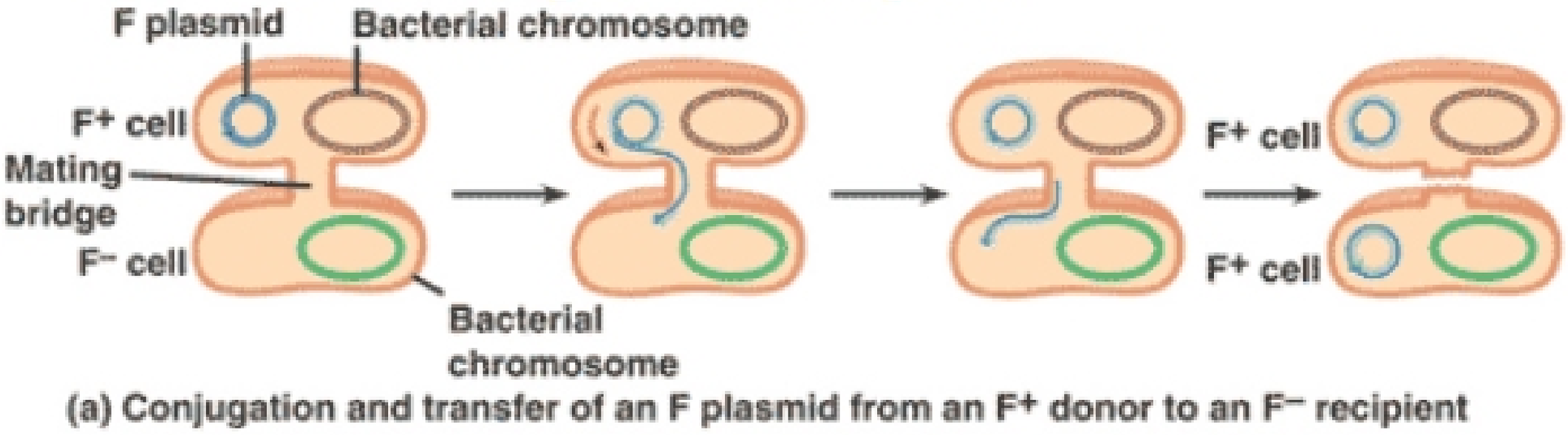
8.1 Bacteria Mutations and Cultures

- Bacteria are haploid, all mutations are expressed in offspring
- Mutant cells can be isolated and established independently from parent strain
- Bacteria are grown in a liquid culture medium or in a petri dish on semisolid agar
- **Minimal medium:** organic carbon source and a variety of ions (Na+, K+, Ca+, Mg 2+, NH4+) used to show WILD TYPE microorganisms
    - **Prototroph** is a bacterium that is able to synthesize all essential organic compounds.
    - **Auxotroph** is a bacterium that has lost ability to synthesize one or more organic components through a mutation.
    - Ex: if a bacteria loses ability to make amino acid histamine then its called *his-*
- To study bacteria quantitatively we use liquid culture mediums
- **Serial dilutions** is a technique used when the number of colonies is too great by using a series of successive dilutions of the original liquid culture until the number of colonies is reduced
    - *Since each colony arose from a single bacterium*, the number of colonies multiplied by the dilution factor represents the number of bacteria in each milliliter of the initial inoculum used to start the dilution series

8.2 Genetic Recombination

- Recombination is the replacement of one or more genes present in the chromosome of o one cell, alters the genotype
- 3 processes that result in the transfer of genetic info in prokaryotes: **conjugation, transformation, and transduction**
- **Vertical gene transfer:** when a transfer of genetic info occurs between same species
- **Horizontal gene transfer:** when members of distinct but related species exchange genetic info
- Conjugation
    - Process by which genetic info from one bacteria is transferred to and recombined with another
    - Lederberg and Tatum experiment, two multiple autotrophs of E. Coli strain K12
        - Strain A: required methionine (met) and biotin (bio) in order to grow
        - Strain B: required threonine (thr), leucine (leu), and thiamine (thi)
        - Grown separately in supplemented media then mixed together
        - Assumed that any prototroph recovered must have arisen as a result of some genetic exchange and recombination between the mutant strains
        - F+ cells were chromosome donors whereas the recipient cell was designated F-
        - F+ crossed with a F- yields F+
    - CELL CONTACT IS NECESSARY

- Physical interaction is established with the **f pilus or sex pilus** which is a tubular extension of the cell
- **Fertility factor** confers the ability to donate part of their chromosome during conjugation, is a mobile element
  - Transfer of f factor involves separation of two strands of DNA and movement of one strand into the recipient
  - Autosomal genetic unit called a **plasmid**



(a) Conjugation and transfer of an F plasmid from an F+ donor to an F− recipient

- An **Hfr (high-frequency recombination)** strain behaves as donor and is a special class of F^+ cells.
- An Hfr strain can donate genetic information to an F^- cell, but the recipient does not become F^+.
  - $F^+ \times F^- \rightarrow$ recipient becomes F^+ (low rate of recombination).
  - Hfr x $F^- \rightarrow$ recipient remains F^- (high rate of recombination).
- Interrupted matings demonstrated that specific genes in an Hfr strain are transferred and recombined sooner than others
- An ordered linear transfer of genes is correlated with the length of time conjugation proceeded
  - The gene order and distance between genes could be predicted
  - Basis for first genetic map in bacteria
- Gene transfer by Hfr strains led to the understanding that the *E. coli* chromosome is circular
- During conjugation between an Hfr and an F^- cell, the position of the F factor determines the initial point of transfer
  - –Conjugation rarely allows the entire chromosome to pass across the conjugation tube.
- This procedure has established the location of approximately 1000 genes

8.3 Rec Proteins

- How does donor DNA replace a homologous region on the recipient chromosome? Major discoveries were made by isolating a group of mutations that impaired the recombination process
- Researchers isolated several functional gene products in normal cells that were missing in rec mutant cells
  - The first product was **RecA protein**, plays an important role in recombination of single stranded DNA or the linear end double stranded DNA that has unwound.

- **Single stranded displacement** is a common form of recombination
- 2nd gene product was RecBCD protein, an enzyme consisting of polypeptide subunits encoded by three other rec genes, unwinds double helix and facilitates recombination

8.4 Plasmids

- Extra chromosomal hereditary unit in a closed circle form are **plasmids**
  - Contain one or more genes
  - Replication depends on the same enzymes that replicate the chromosome on the host cell
  - Classified according to the genetic info specified by their DNA
  - Distributed to daughter cells and exist in multiple copies in the cytoplasm
- **R plasmids** consist of two components:
  - The **resistance transfer factor** (**RTF**) and **r-determinants**
  - RTF encodes genetic information essential to transferring the plasmid between bacteria.
  - R-determinants confer resistance to antibiotics
- **Col Plasmid** is distinct from the R plasmid. It encodes one or more proteins that are highly toxic to bacterial strains that do not harbor the same plasmid.
  - These proteins are called **colicins** and can kill neighboring bacteria

8.5 Transformation

- **Transformation** is another mechanism for recombining genetic information in some bacteria
- Small pieces of extracellular DNA are taken up by the living bacteria, potentially leading to a *stable change in the genes* of the recipient cell.
- Transformation process has many steps divided into two categories: (1) entry of DNA into recipient cell (2) and recombination of the donor DNA with its homologous region in the recipient chromosome
- Only cells in the state of **competence** can take up DNA, entry is through a limited number of receptor sites
- After entry, one strand of DNA is digested by nucleases leaving one strand to participate in transformation. Once it is integrated into the host strand, the helical region where both strands exist (from different sources) is called the **heteroduplex**.
- In prokaryotes, linkage refers to the proximity of genes that permits cotransformation

8.6 Bacteriophages and Bacterial Viruses

- Transduction is understood through the genetics of bacteriophages which must undergo recombination themselves
- **Bacteriophages** (or phages) are viruses that can infect a host bacterium by injecting their DNA.
- Bacteriophage T4 belongs to a group of related bacterial viruses called T-even viruses.