Genetic Transfer & Recombination in Bacteria

- <u>Genetic recombination</u>: transfer of DNA from one organism (donor) to another recipient. The transferred donor DNA may then be integrated into the recipient's nucleoid by various mechanisms (homologous, non-homologous).
- <u>Homologous recombination</u>: homologous DNA sequences having nearly the same nucleotide sequences are exchanged by means of Rec A proteins. This involves breakage and reunion of paired DNA segments as seen in Natural mechanisms of genetic recombination in bacteria include:
 - a. Transformation
 - b. Transduction
 - c. Conjugation

Prokaryotic organisms often donate genetic information (DNA) to recipient cells, which may acquire new characteristics as a result. These characteristics may increase the survivability of a species in a changing environment. They improve the chances that the population will contain individuals suited to the new conditions who can repopulate the environment with their similarity adapted progeny.

Some of these genetic transfers, such as the transposon exchanges mentioned above, have a profound impact on the outcome of infectious disease in humans, especially if the new environmental condition is the presence of antibiotics in an infected body.

Gene transfers have also provided the basis for genetic engineering.

Genes are transferred among bacteria in one direction-from donor to recipient. In most cases, only part of the DNA is transferred. Once this DNA fragment is in the recipient, it may recombine by breakage of the host chromosome and union of the free ends with the newly received DNA fragment. Recombination, therefore, results in the stable incorporation of the new genes into the recipient's chromosome.

In one type of recombination, the new DNA replaces an equivalent portion of the recipient's DNA, which is "discarded". The new DNA segment is homologous with replaced segment that is, it contains genes for the same functions. The recipient cell, however, may receive a different version of a gene, thereby acquiring a new trait from the donor cell's DNA.

For example, a mutant, non-functional gene for capsule formation may be replaced by a functional wild-type gene from the donor. The appearance of one of the donor's traits in a recipient cell suggests that genetic recombination has occurred.

Plasmids and transposons that integrate do not always require homologous regions to insert themselves into DNA. All they require is a particular recognition sequence that specifies a point at which they can integrate into the chromosome.

In these cases, none of the recipient's DNA is lost, although gene function may be destroyed if the new DNA is inserted into the middle of a gene. In addition to recognition sequences, the transferred DNA element must be compatible with the recipient cell for the genetic information to be maintained and expressed.

Transformation

- ➤ The transfer of genetic information by free, extracellular DNA is called transformation.
- Genetic recombination in which a DNA fragment from a <u>dead</u>, <u>degraded</u> <u>bacterium</u> enters a <u>competent</u> recipient bacterium and it is exchanged for a piece of the recipient's DNA.
- Involves 4 steps

Transformation may be an important natural mechanism of genetic transfer among bacteria. It occurs among streptocci and bacilli and certain gram-negative organisms, notably *Neisseria* and *Haemophilus*. These cells, however, are not always capable of being recipients in transformation.



Competence

- Competence is the ability of a bacterial species or strain to take up DNA from its environment.
- Many species are naturally competent, such as *Streptococcus pneumioniae*, *Acinetobacter calcoaceticus*, *Neiserria gonorrheae*, *and Bacillus subtilis*.
- Naturally competent species possess a nucleic acid transporter that spans their cell wall & plasma membrane.

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- ➤ The transporter binds to double-stranded DNA, hydrolyzes one of the strands, and pulls the other strand into the recipient cell.
- The donor DNA strand may then recombine with the recipient chromosome, possibly changing the phenotype of the recipient to the donor phenotype.
- Competence can be induced in some species that are not naturally competent
 - In certain noncompetent gram-negative species (for example, *Escherichia coli*), competence can be induced by treating the cells with divalent calcium ions (Ca2+), usually as a solution of calcium chloride.
 - In certain noncompetent gram-positive species (for example, *Geobacillus stearothermophilus*), competence can be induced by "protoplasting," or removing the cell wall from the cells by lysozyme digestion.

Transduction

Genetic recombination in which a DNA fragment is transferred from one bacterium to another by a bacteriophage.







Contraction of the tail sheath of T4

What are Bacteriophages?

Bacteriophage (phage) are obligate intracellular parasites that multiply inside bacteria by making use of some or all of the host biosynthetic machinery (i.e., viruses that infect bacteria).

- There are two types of transductions:
 - Generalized transduction: A DNA fragment is transferred from one bacterium to another by a <u>lytic bacteriophage</u> that is now carrying donor bacterial DNA due to an error in maturation during the lytic life cycle.
 - **Specialized transduction**: A DNA fragment is transferred from one bacterium to another by a <u>temperate bacteriophage</u> that is now carrying donor bacterial DNA due to an error in spontaneous induction during the lysogenic life cycle.

Seven steps in Generalised Transduction

- **1.** A <u>lytic bacteriophage</u> adsorbs to a susceptible bacterium.
- 2. The bacteriophage genome enters the bacterium. The genome directs the bacterium's metabolic machinery to manufacture bacteriophage components and enzymes.
- **3.** Occasionally, a bacteriophage head or capsid assembles around a fragment of donor bacterium's nucleoid or around a plasmid instead of a phage genome by mistake.
- 4. The bacteriophages are released.
- 5. The bacteriophage carrying the donor bacterium's DNA adsorbs to a recipient bacterium.
- 6. The bacteriophage inserts the donor bacterium's DNA it is carrying into the recipient bacterium.
- 7. The donor bacterium's DNA is exchanged for some of the recipient's DNA.



recipient bacterium's nucleoid

Six steps in Specialised Transduction

- **1.** A temperate bacteriophage adsorbs to a susceptible bacterium and injects its genome.
- 2. The bacteriophage inserts its genome into the bacterium's nucleoid to become a prophage.
- 3. Occasionally during spontaneous induction, a small piece of the donor bacterium's DNA is picked up as part of the phage's genome in place of some of the phage DNA which remains in the bacterium's nucleoid.
- 4. As the bacteriophage replicates, the segment of bacterial DNA replicates as part of the phage's genome. Every phage now carries that segment of bacterial DNA.
- 5. The bacteriophage adsorbs to a recipient bacterium and injects its genome.
- 6. The bacteriophage genome carrying the donor bacterial DNA inserts into the recipient bacterium's nucleoid.



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Bacterial Conjugation

Bacterial Conjugation is genetic recombination in which there is a transfer of DNA from a living donor bacterium to a recipient bacterium. Often involves a sex pilus.

The 3 conjugative processes

- I. F^+ conjugation
- **II.** Hfr conjugation
- III. Resistance plasmid conjugation

I. F⁺ Conjugation Process

<u>**F**</u>⁺<u>**Conjugation:-**</u> Genetic recombination in which there is a transfer of an \overline{F} ⁺ plasmid (coding only for a sex pilus) but not chromosomal DNA from a male donor bacterium to a female recipient bacterium. Involves a sex (conjugation) pilus. Other plasmids present in the cytoplasm of the bacterium, such as those coding for antibiotic resistance, may also be transferred during this process.

The 4 stepped F⁺ Conjugation

- The F⁺ male has an F⁺ plasmid coding for a sex pilus and can serve as a genetic donor.
- The sex pilus adheres to an F⁻ female (recipient). One strand of the F⁺ plasmid breaks.
- The sex pilus retracts and a bridge is created between the two bacteria. One strand of the F⁺ plasmid enters the recipient bacterium.
- Both bacteria make a complementary strand of the F⁺ plasmid and both are now F⁺ males capable of producing a sex pilus. There was no transfer of donor chromosomal DNA although other plasmids the donor bacterium carries may also be transferred during F⁺ conjugation.





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II. Hfr Conjugation

Genetic recombination in which fragments of chromosomal DNA from a male donor bacterium are transferred to a female recipient bacterium following insertion of an F^+ plasmid into the nucleoid of the donor bacterium. Involves a sex (conjugation) pilus.

<u>5 stepped Hfr Conjugation</u>

- An F⁺ plasmid inserts into the donor bacterium's nucleoid to form an Hfr male.
- The sex pilus adheres to an F⁻ female (recipient). One donor DNA strand breaks in the middle of the inserted F⁺ plasmid.
- 3. The sex pilus retracts and a bridge forms between the two bacteria. One donor DNA strand begins to enter the recipient bacterium. The two cells break apart easily so the only a portion of the donor's DNA strand is usually transferred to the recipient bacterium.
- 4. The donor bacterium makes a complimentary copy of the remaining DNA strand and remains an Hfr male. The recipient bacterium makes a complementary strand of the transferred donor DNA.
- 5. The donor DNA fragment undergoes genetic exchange with the recipient bacterium's DNA. Since there was transfer of some donor chromosomal DNA but usually not a complete F⁺ plasmid, the recipient bacterium usually remains F⁻.



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III. Resistant Plasmid Conjugation

Genetic recombination in which there is a transfer of an R plasmid (a plasmid coding for multiple antibiotic resistance and often a sex pilus) from a male donor bacterium to a female recipient bacterium. Involves a sex (conjugation) pilus.

4 stepped Resistant Plasmid Conjugation

- 1. The bacterium with an R-plasmid is multiple antibiotic resistant and can produce a sex pilus (serve as a genetic donor).
- The sex pilus adheres to an F⁻ female (recipient). One strand of the Rplasmid breaks.
- 3. The sex pilus retracts and a bridge is created between the two bacteria. One strand of the R-plasmid enters the recipient bacterium.
- **4.** Both bacteria make a complementary strand of the R-plasmid and both are now multiple antibiotic resistant and capable of producing a sex pilus.

Conjugation in Gram-positive bacteria.



Conjugation among gram-positive bacteria is neither mediated by pili nor driven by an F plasmid. Donor bacteria do, however, contain conjugative plasmids that mediate conjugation between donor and recipient bacteria, during which a copy of plasmid DNA is transferred. Genetic information contained in the donor's plasmid directs the synthesis of a surface substance that promotes binding to recipient cells. This binding substance is produced only in the presence of recipient cells. In some cases, the recipient releases a chemical that induces the plasmid's binding substance gene.