PHARMACEUTICAL MICROBIOLOGY and IMMUNOLOGY

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OBJECTIVES

Genetic Material Transfer in Bacteria

- Transformation
- Conjugation
- Transduction

Transformation

In bacteria genetic material transfer happens in three ways:

Transformation:

Some bacteria have ability to uptake **naked DNA fragment from the surrounding environment**. When such a DNA confers new property to the bacterium, it is termed transformation

Transduction

Transfer of genetic material **through mediation of bacteriophage** is known as transduction

Conjugation

- Transfer of genetic material from one bacterium to another **through the mediation of sex pili** is known as conjugation
- Any property that is coded on a transmissible plasmid can be transferred to a recipient bacterium
- Properties such as drug resistance mediated by beta-lactamases, bacteriocin production can be transferred by conjugation

Genetic Material Transfer in Bacteria

- **Genetic recombination** refers to the exchange of the genes between two DNA molecules to form new recombinations of genes

- DNA is always transferred **from a donor to a recipient**


Movie
time

Transformation

- In 1928, a British bacteriologist named Frederick Griffith first demonstrated bacterial transformation

- *Streptococcus pneumoniae*'s pathogen strains are cover by a polysaccharide capsule that protects bacteria from the host's immune system

  "S" strain is smooth, encapsulated, deadly

  "R" strain is rough, unencapsulated, harmless

Transformation-Griffith's Experiment

A) Injected mice with the **'R' strain of bacteria**
Result: no sickness or death

B) Injected mice with the **'S' strain of bacteria**
Result: dead mice

C) Injected mice with **heat-killed 'S' strain** of bacteria
Result: no sickness or death

Transformation - Griffith's Experiment

D) Injected mice with a mixture of **harmless "R" strain** bacteria and **heat-killed "S" strain** bacteria
Result: dead mice

In Griffith's experiment, segments of the genetic information from the "S" strain bacteria became "naked" when the cells were heat-ruptured

It is observed by this experiment that the gene that formed the capsule was released from the dead pneumonia and transferred to the R strains

Transformation

- In order for bacterial transformation to occur, there needs to be:

  – **"naked DNA":** DNA not found in a cell ("extracellular" DNA)

  – **"competent" bacterial cells:** ability to take up extracellular DNA

Transformation

- **“Competent”** bacteria cells take up DNA segments through their cell membranes

- Some bacteria cells are naturally “competent”
- Some bacteria cells can become artificially “competent” through chemicals, heat-shocking and electro-shocking

Competent: capable of taking up pure DNA from the external medium

Conjugation

- **Conjugation** occurs when plasmid DNA is transferred **from donor to recipient bacterium by direct contact via a sex pilus**
- In bacteria, the DNA transfer is one way
- A donor cell attaches to a recipient by a pilus, pulls it closer and transfers DNA
- A piece of DNA called the F factor is required for the production of pili

Conjugation

- Bacteria that have **F factor (fertility-sex factor)** are called **male (F+ bacteria)**, the ones that **do not have are called female (F- bacteria)**
- Cells containing the F plasmid function as DNA donors during conjugation
- Cells without the F factor function as DNA recipients during conjugation
- The F factor is transferable during conjugation

Conjugation

- **F plasmid** carries a **tra** and **trb** locus, consist of about 40 genes
- The tra locus includes the pilin gene and regulatory genes, which together form pili on the cell surface
- The locus also includes the genes for the proteins that attach themselves to the surface of F(−) bacteria and initiate conjugation

Conjugation

- Tra (transfer) genes control the transfer of genetic material from bacteria to bacteria
- These genes are regulated inside an operon
- Conjugation starts with the creation of sex pilus in the bacteria under the control of some genes in this operon

Conjugation

- Sex pilus which exists only in F(+) bacteria plays an important role in cell-to-cell contact of two bacteria during conjugation and the transfer of genetic material between cells

- Sex pilus shows antigenic specificity **(F antigen)**. The F pilus may be detected on the surface of both F(+) and Hfr cells by the presence of the F antigen

Conjugation

- **F factor** is an extrachromosomal factor (plasmid). In some cases, F factor attaches to the main bacteria chromosome and integrated into the DNA of bacteria

- When F factor becomes integrated into the chromosome of a F(+) cell, it makes the cell a **high-frequency recombination cell (Hfr cell)**

Conjugation

- The integrated F factor occasionally leaves the bacterial DNA **(Hfr cell)** and moves back to the cytoplasm
- In some rare cases carrying a few host chromosomal genes along with it
- This type of cells are defined as F prime (F′) cells. F prime cells contain F plasmid carrying some bacterial genes

Conjugation

Some features of Hfr (high-frequency recombination) cells:

- These cells also have sex pilus
- Hfr cells can transfer main chromosome particles of a bacteria to other bacterial cells, especially to F(-) cells and this results in the new recombination
- Hfr bacteria can never be a recipient cell, however they can transfer bacterial chromosome especially to F(-) cells.

Conjugation

- Conjugation can be seen in microorganisms like *E.coli, Salmonella, Shigella, Pseudomonas, Serratia, Vibrio*

- Conjugation may happen between two bacteria that are either the same or different species

PLASMIDS

- A plasmid is a small DNA molecule within a cell that is physically separated from a chromosomal DNA and can replicate independently

- Some plasmids contain **tra genes (transfer genes)** that provide their own transfer from bacteria to bacteria. These are called **conjugative plasmids**

- The ones that do not have these kind of genes and thus can not be transferred to other bacteria on their own are called **non-conjugative plasmids**

PLASMIDS

- Non-conjugative plasmids can be transferred to other bacteria by conjugation only if there are F factors in the bacteria or when they are mobilized via other conjugative plasmids. This is called **plasmid mobilization**

TYPES OF PLASMIDS

- **Fertility Plasmids (F PLASMID)** – carry the fertility genes for conjugation, allow bacteria to mate with each other and transfer of genetic information between two cells

- **Resistance Plasmids (R PLASMID)** – Contain genes that confer resistance against antibiotics and other types of toxins

TYPES OF PLASMIDS

- **Col Plasmids** – contain genes that encode for the antibacterial polypeptides called bacteriocins, which are proteins that kill other bacteria

- **Degradative Plasmids** – Carry genes that enable the bacterium to digest and utilize an unusual substance

- **Virulence Plasmids** – Carry genes that turn a bacterium into a pathogenic strain

Transduction

- Transduction is the process by which **DNA is transferred from one bacterium to another by a bacteriophage**

- Bacteriophages are viruses that infect bacteria

- Bacteriophage can be either lytic or temperate
  - **Lytic** - always lyse (kill) host bacterial cell
  - **Temperate** – can stably infect and coexist within bacterial cell (**lysogeny**) until a lytic phase is induced

Transduction

- There are two types of transduction

  - Specialized Transduction - Generalized Transduction

Specialized Transduction

- Some prophages **integrate into the bacterial genome at a specific location.** When a prophage is induced to lytic phase, it may drag along a piece of the bacterial genome next to the integration site and move that bacterial sequence into the new recipient host cell, changing the recipients genome.

Transduction

- There are two types of transduction

  - Specialized Transduction - Generalized Transduction

Generalized Transduction

When a phage lyses the host bacterial cell, it normally packages phage genome into the capsid. Sometimes the capsid is accidently filled with random pieces of bacterial genome, possibly including plasmids. When the capsid injects the host genes into a new recipient, the new gene can recombine into the recipient genome and cause a change. Virulence and antibiotic resistance genes can be moved by generalized transduction

Specialized 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

Specialized Transduction

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

Generalized Transduction

1. A lytic bacteriophage 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

Generalized Transduction

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