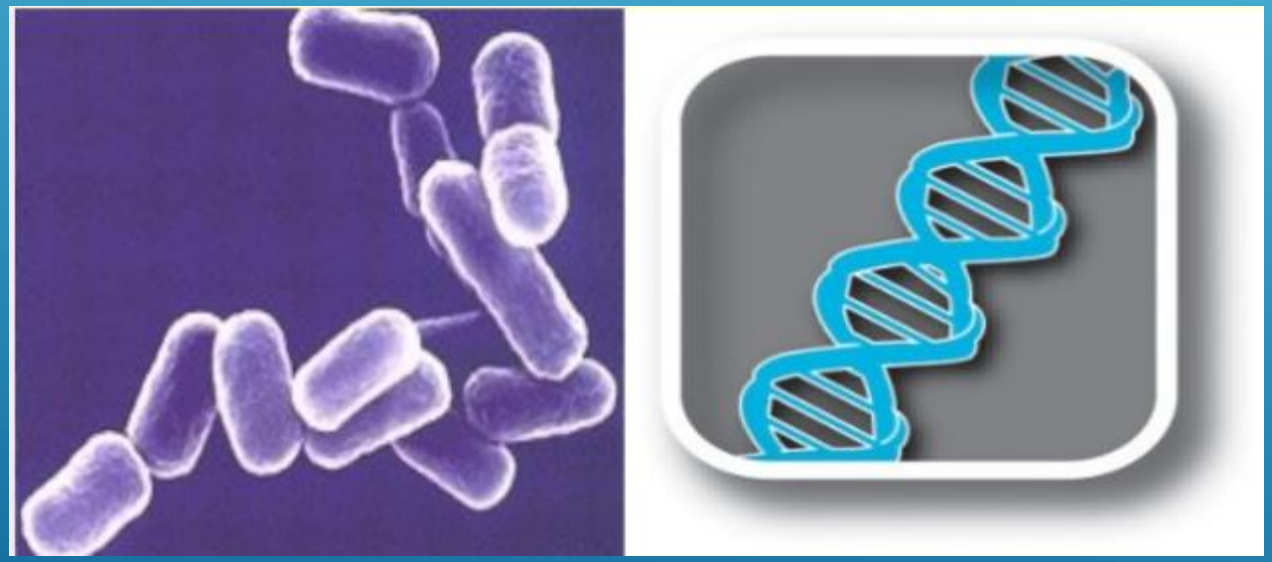
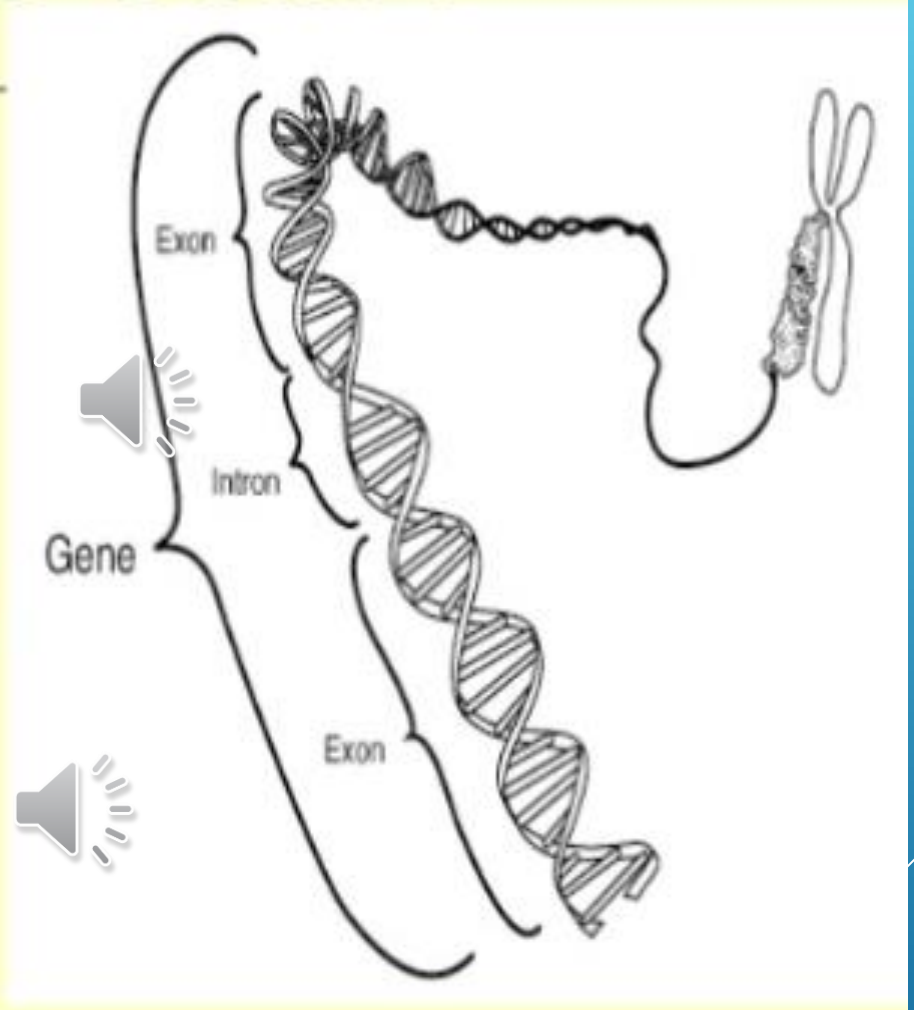
BACTERIAL GENETICS AND GENE TRANSFER



What is a Gene?

- Gene is a **sequence of DNA** carrying **codons** specifying for particular polypeptide.
- DNA contains many Genes(**combinations of hundreds and thousands of Nucleotides**)



Bacterial Chromosome

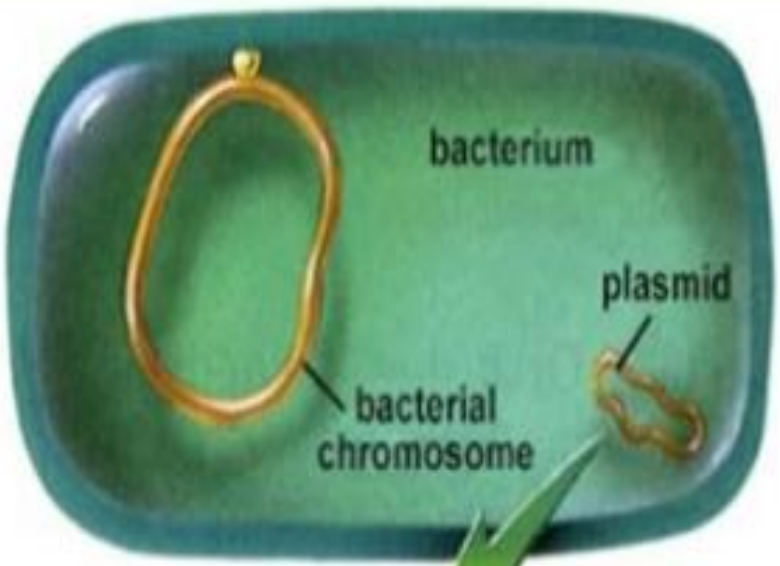
- Contains a **Double stranded** molecules of DNA arranged in circular form.
- Length **1,000 microns**.
- Bacterial DNA contains about **4,000kilobases**
- **1 kb = 1000 base pairs** (A-T) (G-C)

Extra chromosomal Genetic Elements

- Bacteria posses Extra chromosomal genetic elements
- Not Essential for **survival** of Bacteria
- But makes the **Bacteria Resistant to antibiotics**, and makes them survive & also able to produce toxins

Plasmids

- Plasmids are **circular DNA molecules** present in the cytoplasm of the Bacteria
- Their size varies from **1 kbp to over 400 kilobase pairs (kbp).**
- Capable of **Autonomous replication**
- Can transfer **genes from one cell to other**



Plasmids

- Plasmid seem to be ubiquitous in bacteria, May encode genetic information for properties
  1 Resitance to Antibiotics
  2 Bacteriocins production
  3 Enterotoxin production
  4 Enhanced pathogenicity
  5 Reduced Sensitivity to mutagens
  6 Degrade complex organic molecules

by their ability to be transferred to other bacteria

Conjugative

❖ The **sexual transfer** of plasmids to another bacterium through a pilus.

Non-conjugative

❖ Non-conjugative plasmids don't initiate conjugaison. They can only be transferred with the help of conjugative plasmids.

1. **Fertility-(F) plasmids.**

- They are capable of conjugation (they contains the genes for the pili).

2. **Resistance-(R) plasmids**.

- Contain gene (s) that can build resistance against one or several antibiotics or poisons.

3. **Col-plasmids**.

- Contain genes coding for colicines, proteins that can kill other bacteria.

4. Degradative plasmids,

- able to digest unusual substances, e.g., toluene or salicylic acid

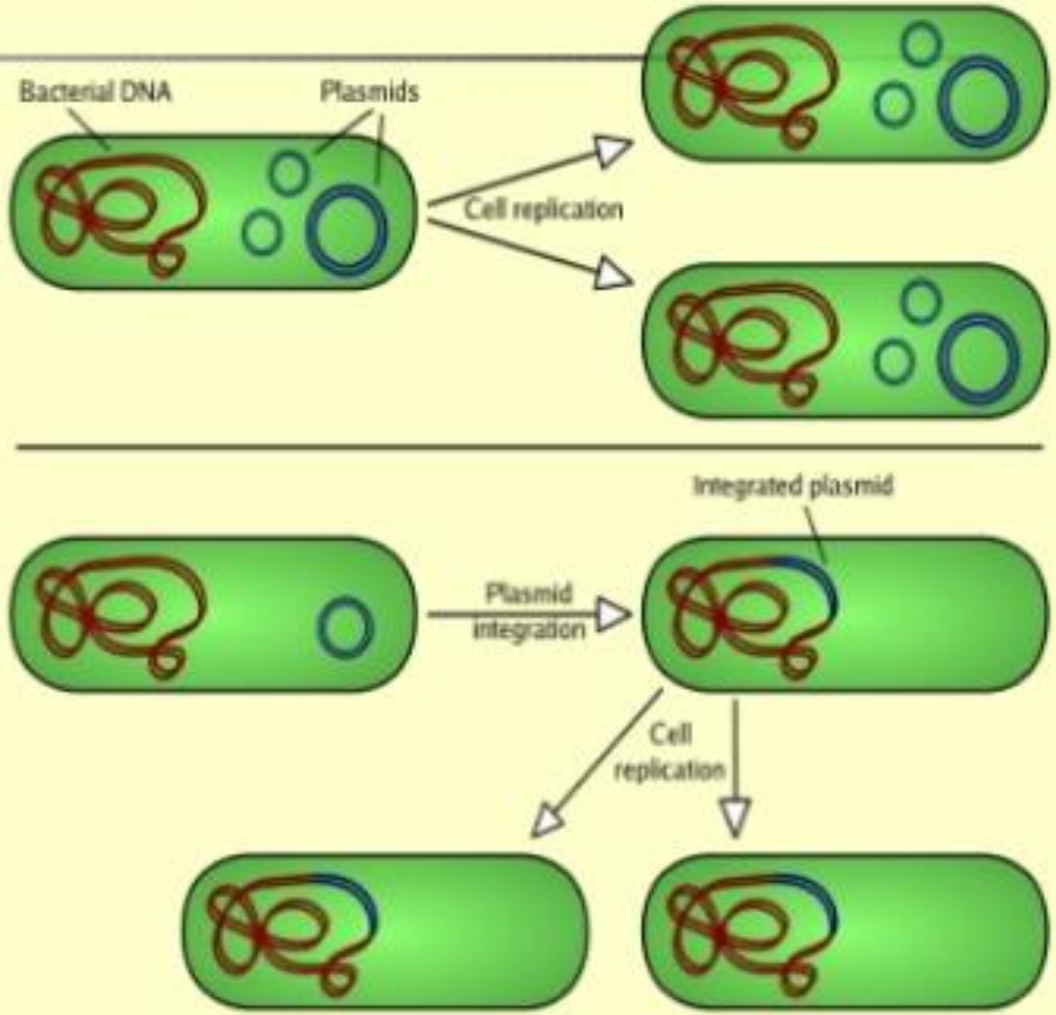
5. Virulence plasmids

- turn a bacterium into a pathogen.

Plasmids

- Can be **integrated** with Chromosomal DNA

- **Episomes**
  -Integrated form of plasmid with DNA



Genotype / Wild Type : Represents all potential genes of bacteria cell.. Its genome.. All Inherited essential biological features & growth patterns.

Phenotype: The expressed genes. The observed characteristics of the individual bacteria species/strain. Expressed by physical & biochemical properties. Growth patterns, Fermentation products, Antibiotic resistance, Toxins production. .etc.

Prokaryotes Vs Eukaryotes Genetics

Prokaryotes	Eukaryotes
Prokaryotes are haploid	**eukaryotes are often diploid**
contain a **single circular chromosome**.	eukaryotes have **linear chromosomes, usually more than 1**
Prokaryotes often contain **"plasmids"**.	Doesnot contain plasmids
In prokaryotes, **translation is coupled to transcription: translation of the new RNA molecule starts before transcription is finished**	**In eukaryotes, transcription of genes in RNA occurs in the nucleus, and translation of that RNA into protein occurs in the cytoplasm**. The two processes are separated from each other.

Gene transfer

In bacteria or other organism ,gene transfer mainly two ways.

1.vertical gene transfer

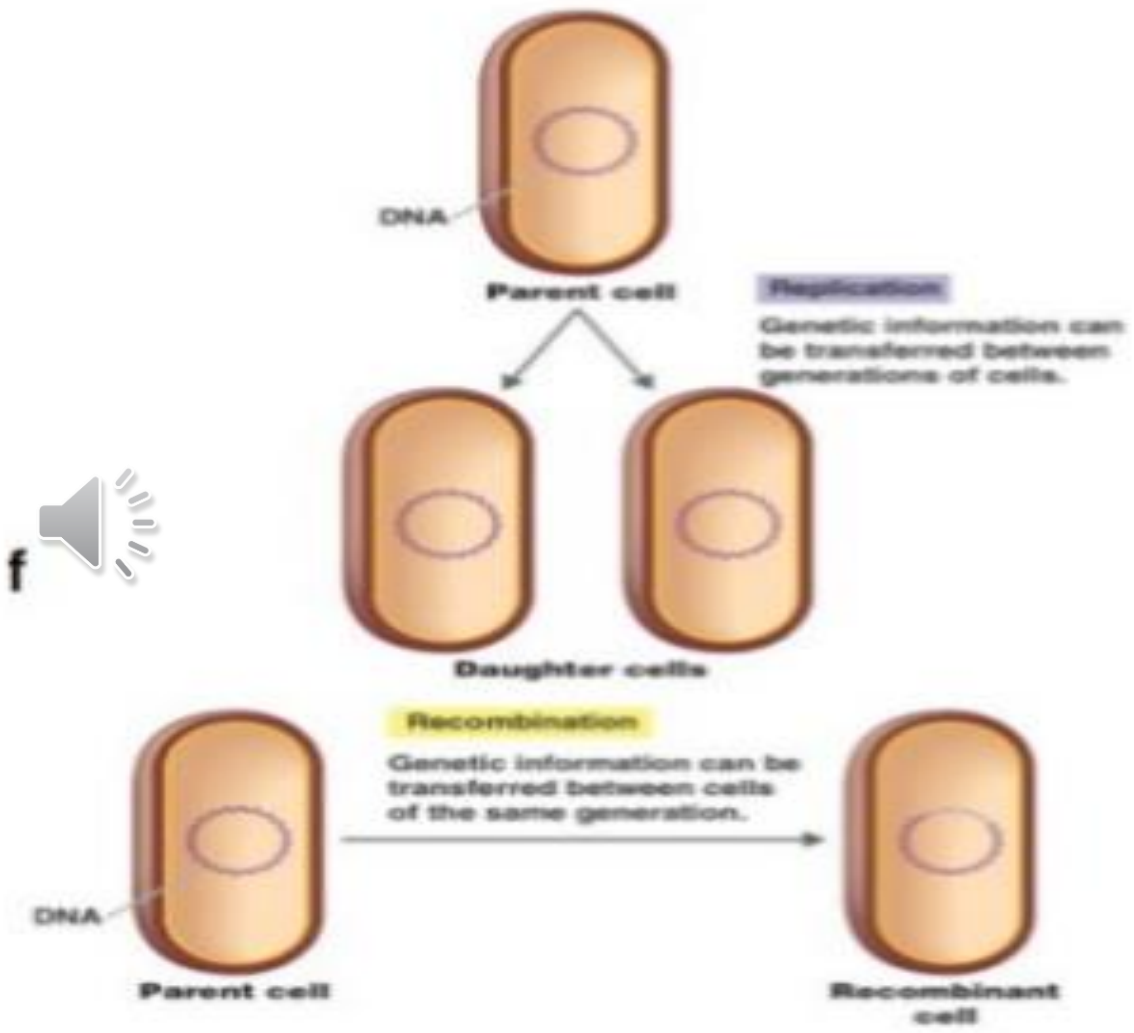
2.horizontal gene transfer

vertical gene transfer

Transfer of gene from mother to daughter cell or parents to ofsprins.

mainlly occurs during the reproduction between generation of cells.

DNA inherited from parental organism



Horizontal gene transfer

Transfer of gene between cells of the same genaration in two different specics.

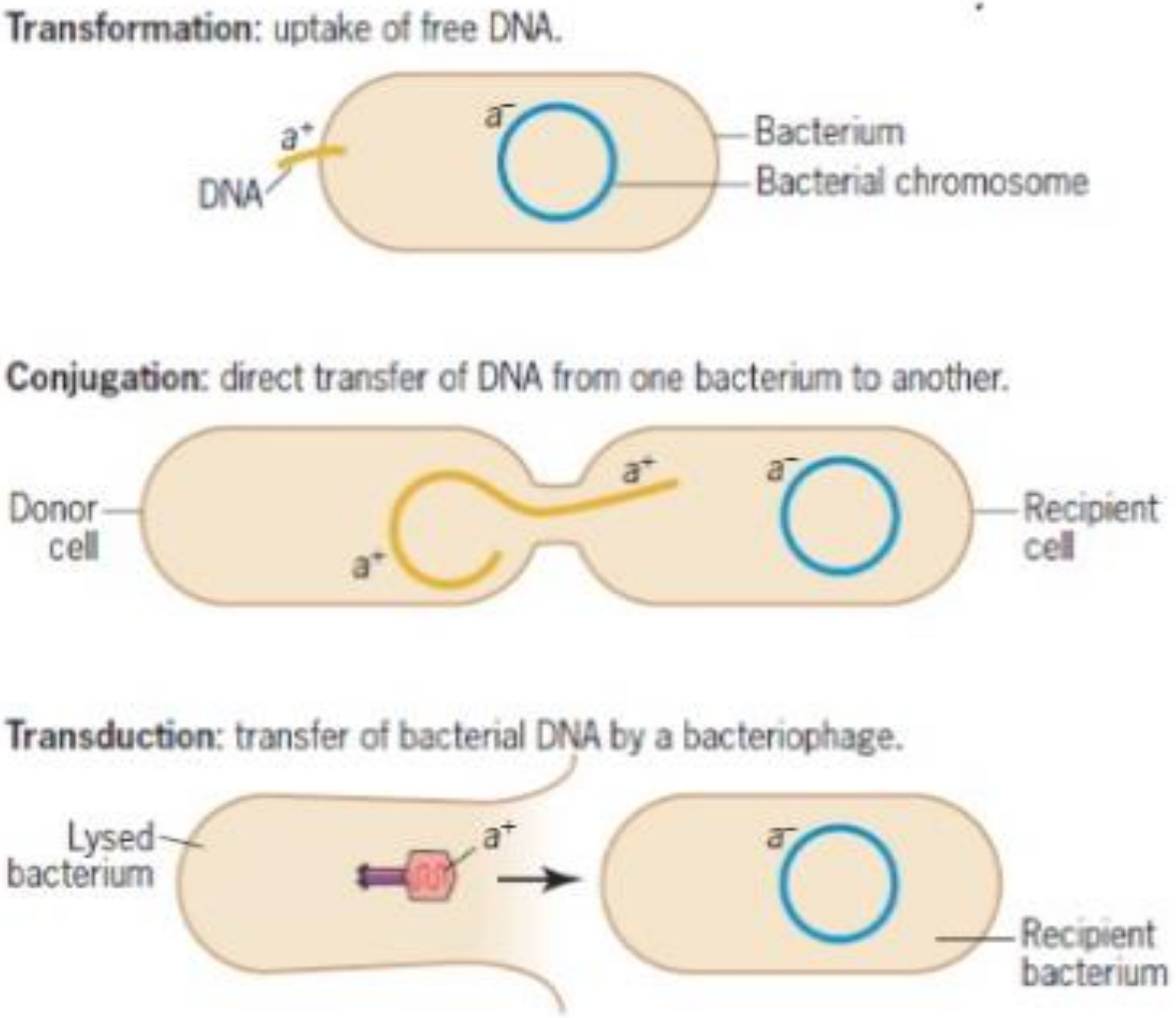
DNA acquired from unrelated individuals

There are three types of horizontal gene transfer.

1.transformation.

2.transduction.

3.conjugation.



■ FIGURE 8.8 The three types of gene transfer in bacteria.

Transmission of Genetic material (Gene Transfer)

- HOW BACTERIA GAIN NEW GENETIC INFORMATION

- MUTATION
- TRANSFORMATION
- CONJUGATION
- TRANSDUCTION

MUTATION

- Is any heritable change in the genetic material
- Mutations may be neutral, beneficial, or harmful
- Mutant- organism or strain whose genome carries a mutation
- Wild type- the usual (native) form of the organism
- Occurrence of mutations:
  - Spontaneous - Occur in the absence of a mutagen
  - Induced - Occur in the presence of a mutagen
- Mutagen: Agent that causes or accelerates rate of mutations

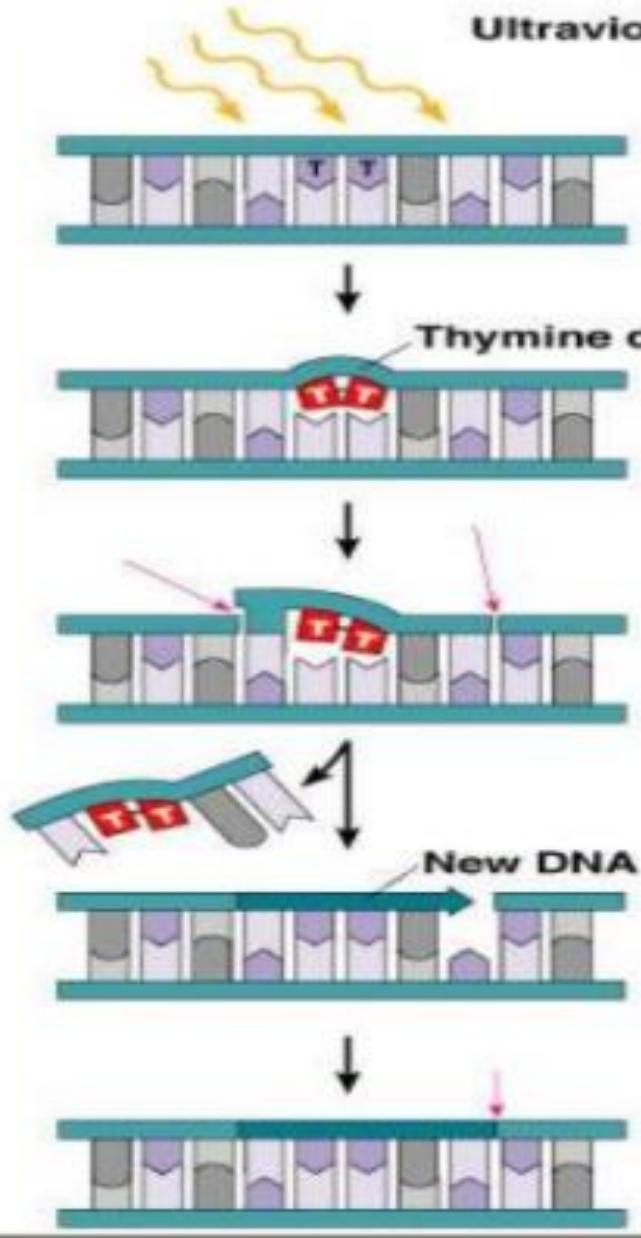
Mutagens

- Mutagens are chemical, physical or biological agents that increase the mutation rate i.e. induce mutations

Physical mutagens

- Ionizing radiation (X rays and gamma rays) causes the formation of ions that can react with nucleotides and the deoxyribose-phosphate backbone
  - Nucleotide excision repairs mutations

- Non ionizing radiation
  - UV radiation causes thymine dimers
  - Light-repair separates thymine dimers

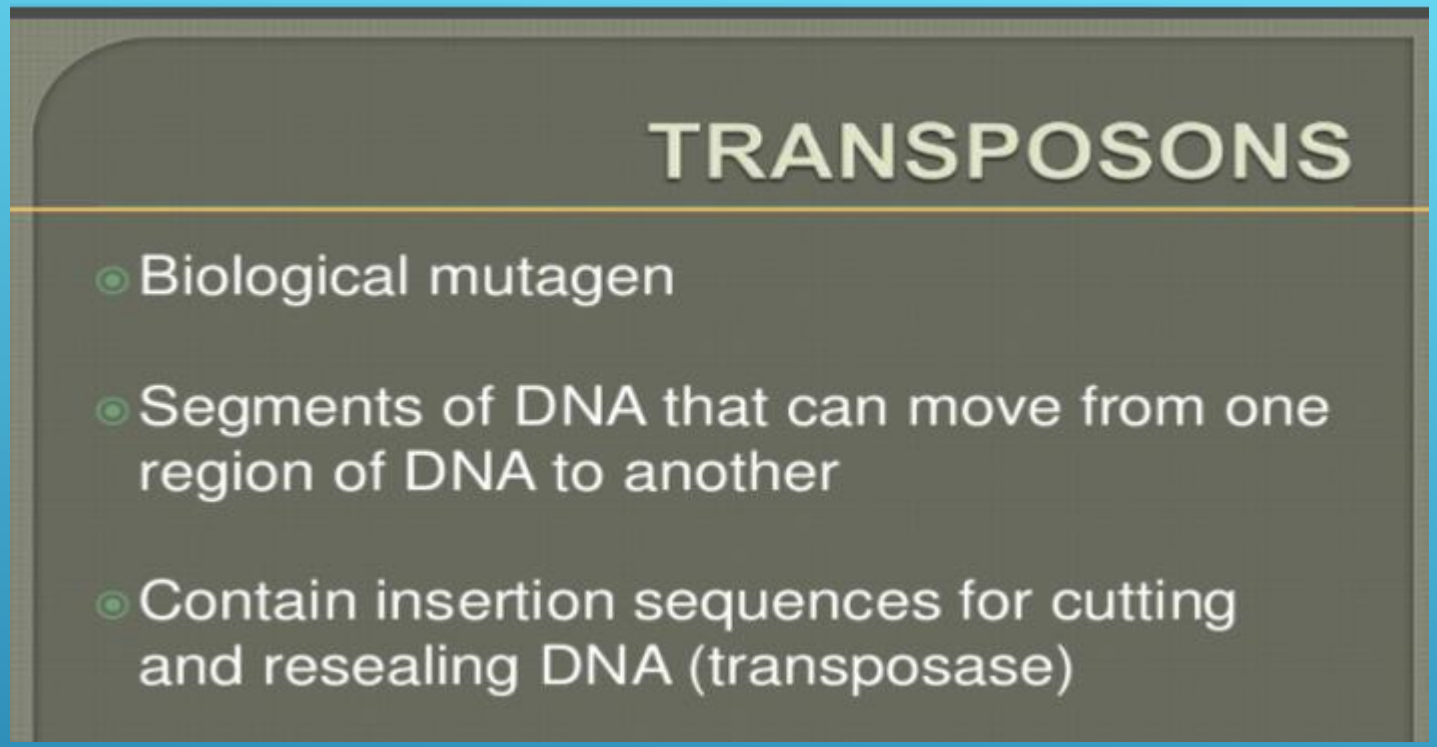


1. Exposure to ultraviolet lighcauses adjacent thymines to become cross-linked, forming a thymine dimer and disrupting their normal base pairing.

2. An enzyme cuts out and removes the damaged DNA.

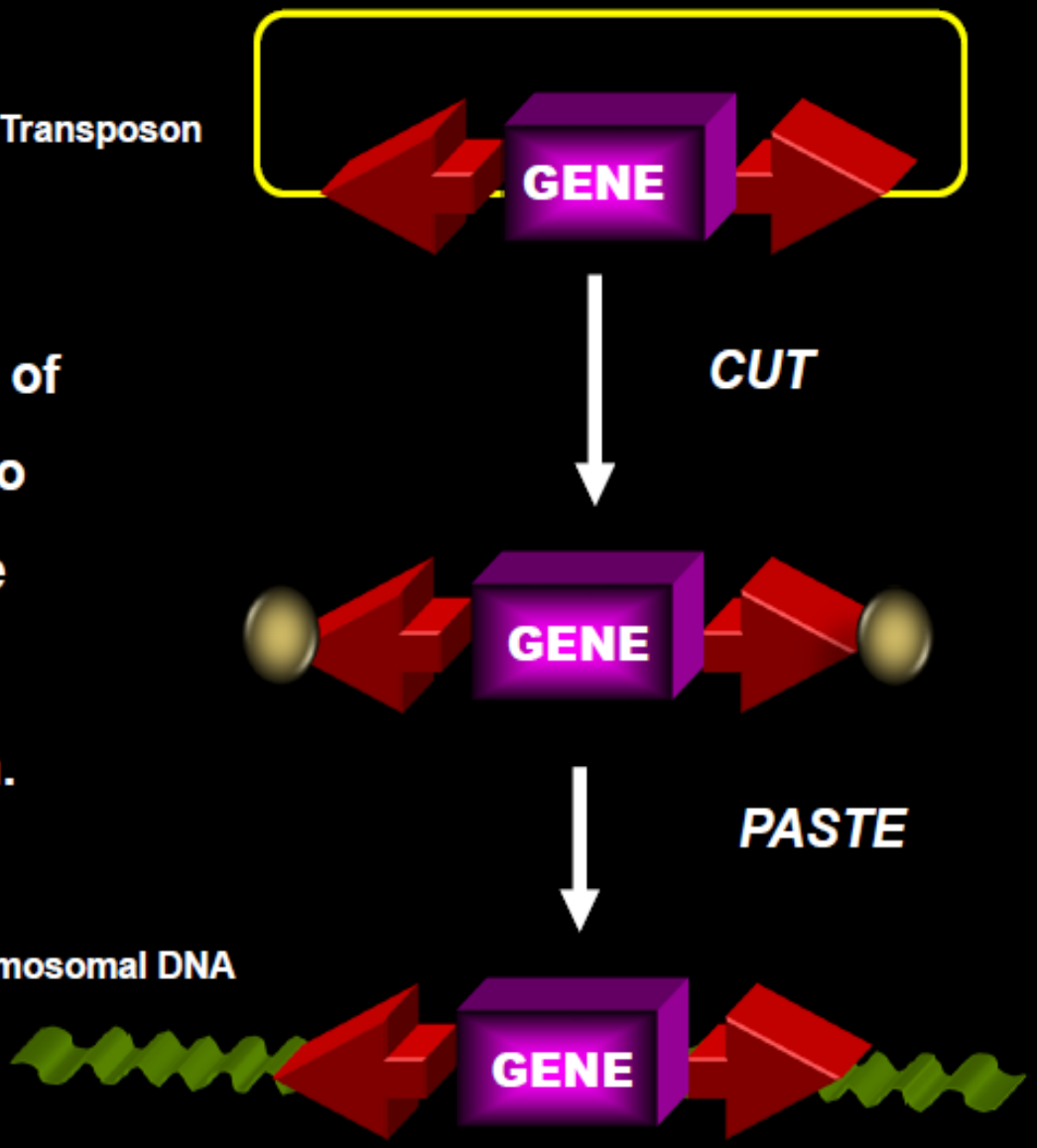
3. DNA polymerase fills the gap by synthesizing new DNA, using the intact strand as a template.

4. DNA ligase seals the remaining gap by joining the old and new DNA.



Jumping genes/copy and paste (Class1) or cut and paste (2)
Between plasmids or between chromosomes and plasmids
•**medical importance** since many **antibiotic resistance genes** are encoded by transposons in antibiotic **resistance plasmids**

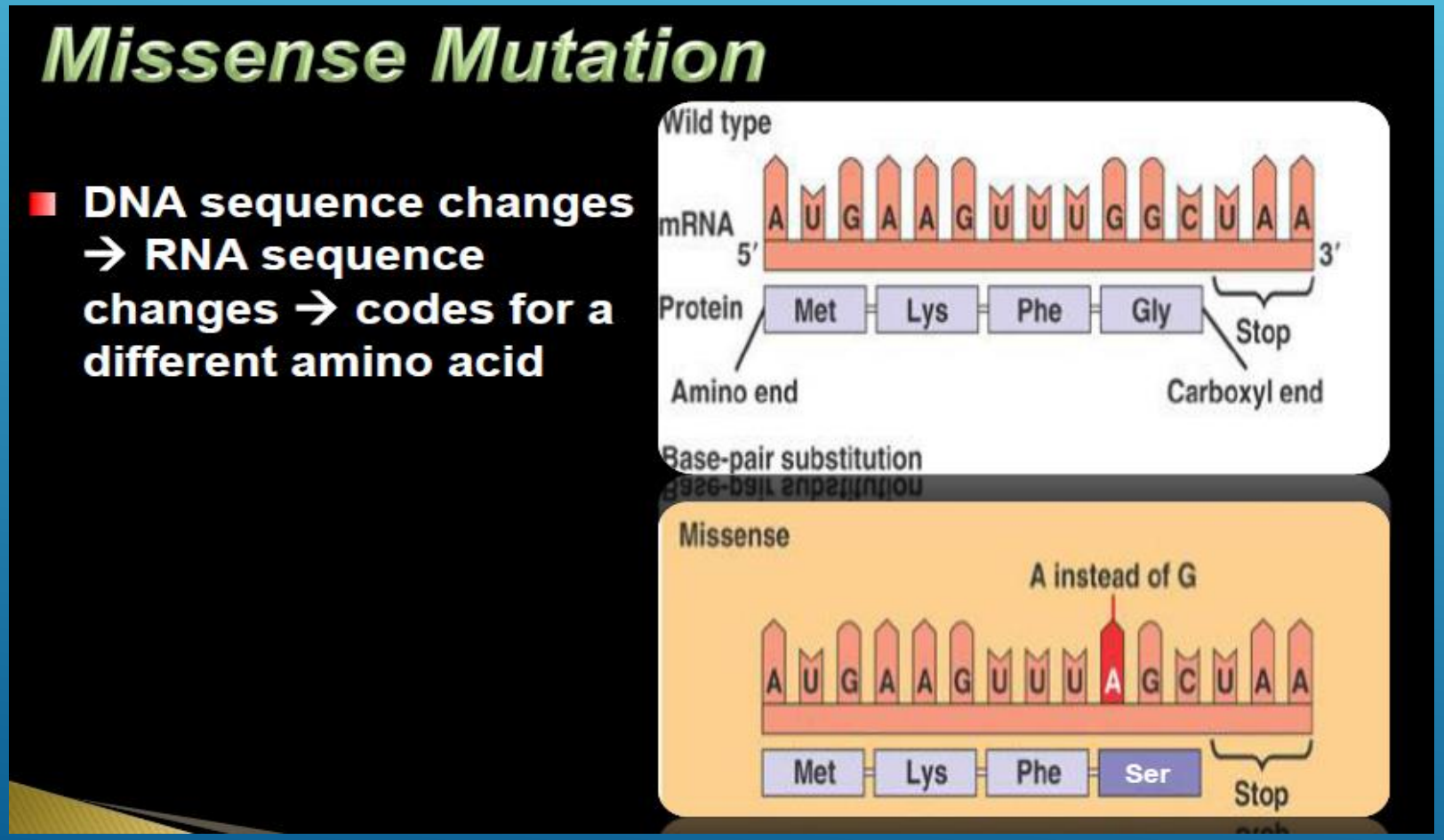
Transposons are sequences of DNA that can move around to different positions within the genome of a single cell, a process called transposition.



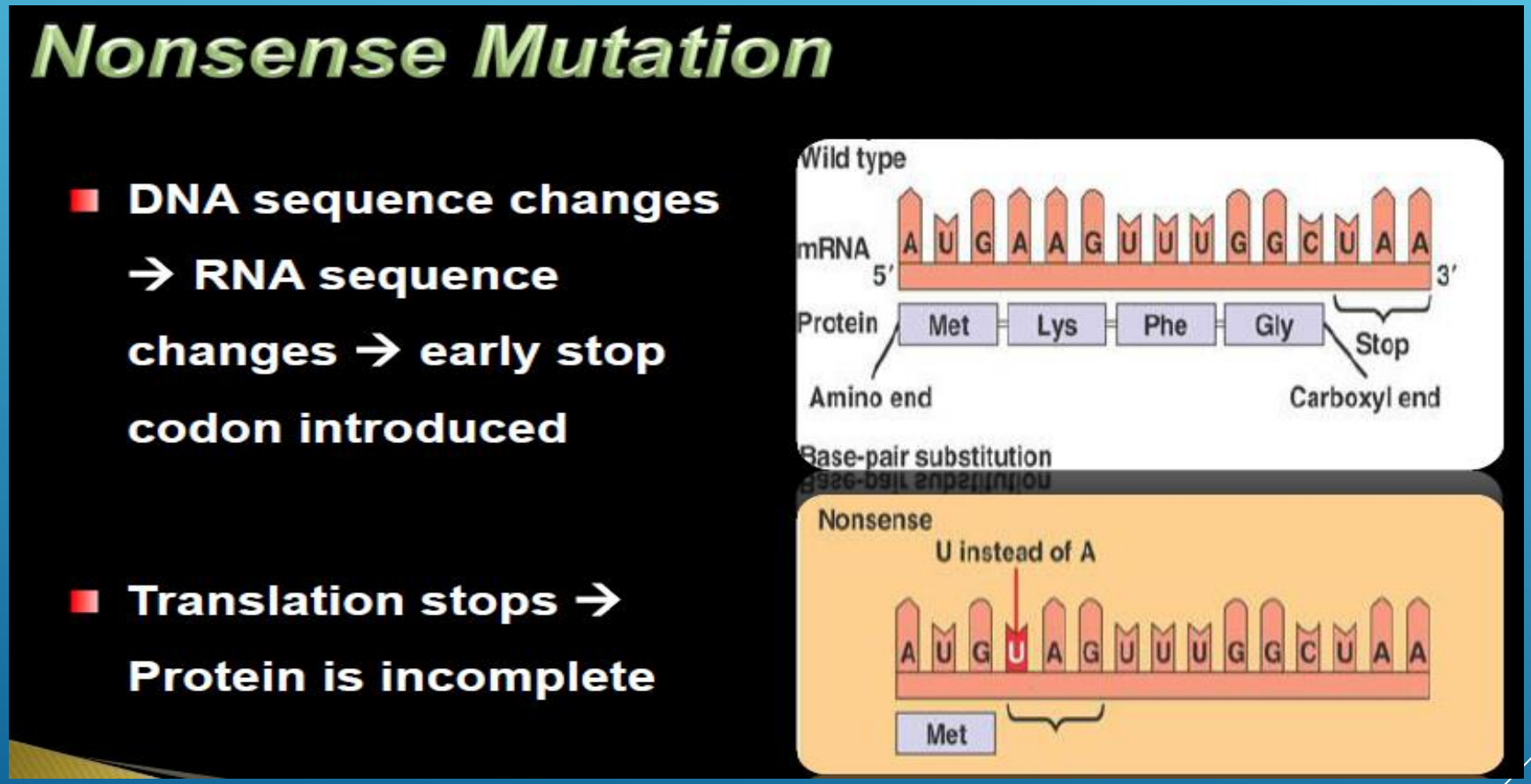
TYPES OF MUTATION

- Base substitution (point mutation)
  - Change in one base
- Missense mutation
  - Result in change in amino acid
- Nonsense mutation
  - Results in a nonsense codon
- Frameshift mutation
  - Insertion or deletion of one or more nucleotide pairs

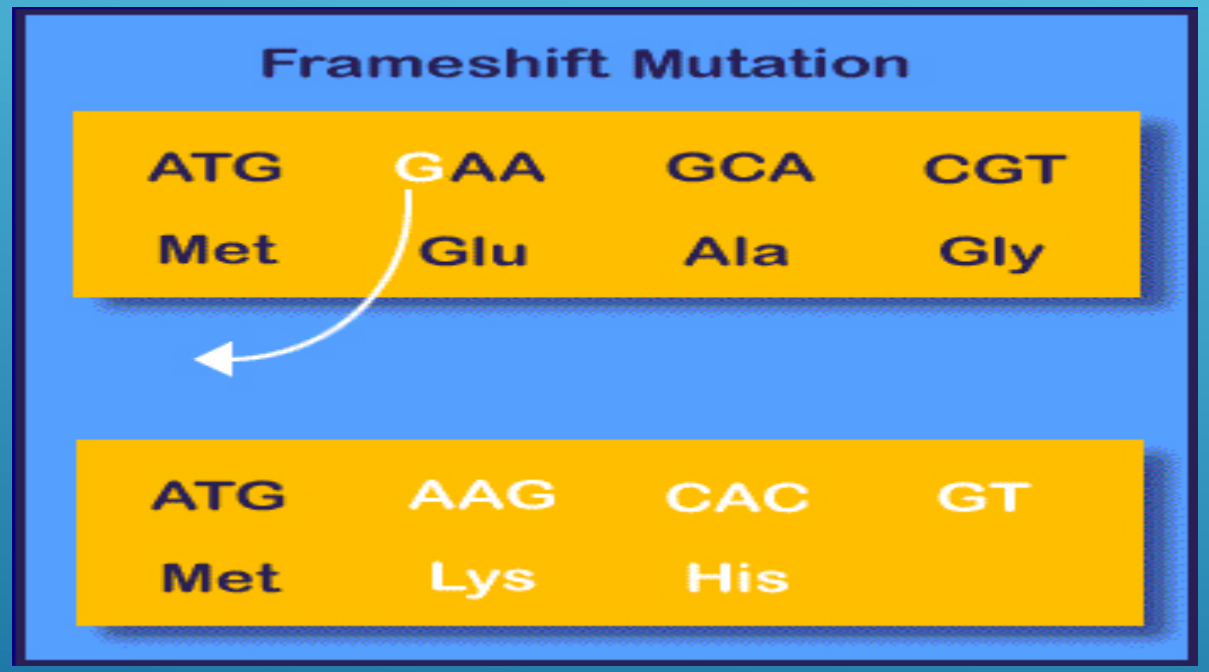
MISSENSE MUTATION: RESULT IN DIFFERENT AMINO ACIDS BEING INSERTED IN PROTEIN



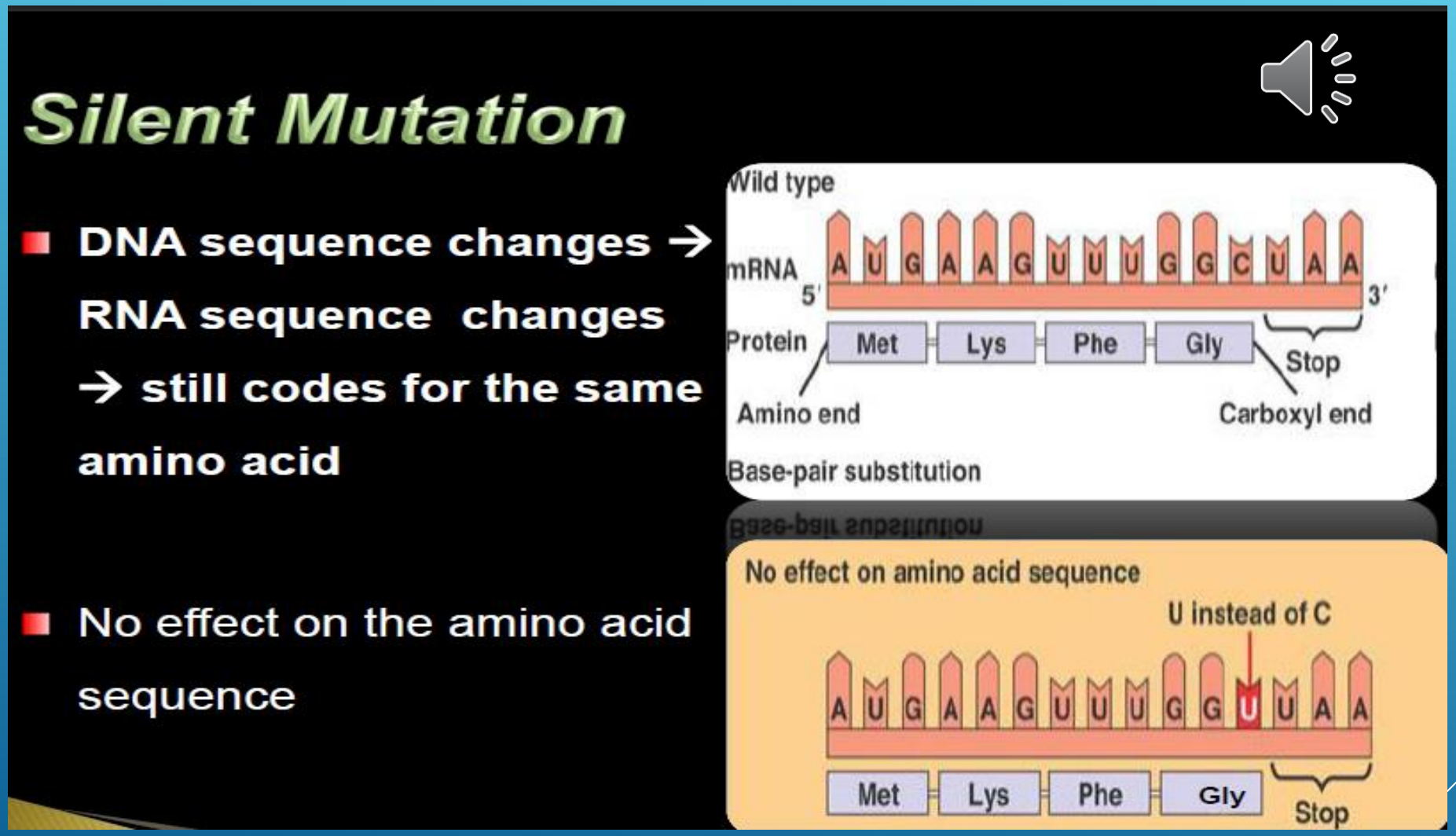
Nonsense mutation: change a codon encoding an amino acid into stop codon that result in failure of protein synthesis



- Frame shift mutation: One or more base are added or deleted, Shift in the reading frame



Silent mutation: the change in the nucleotide sequence doesn't result in a change in protein sequence



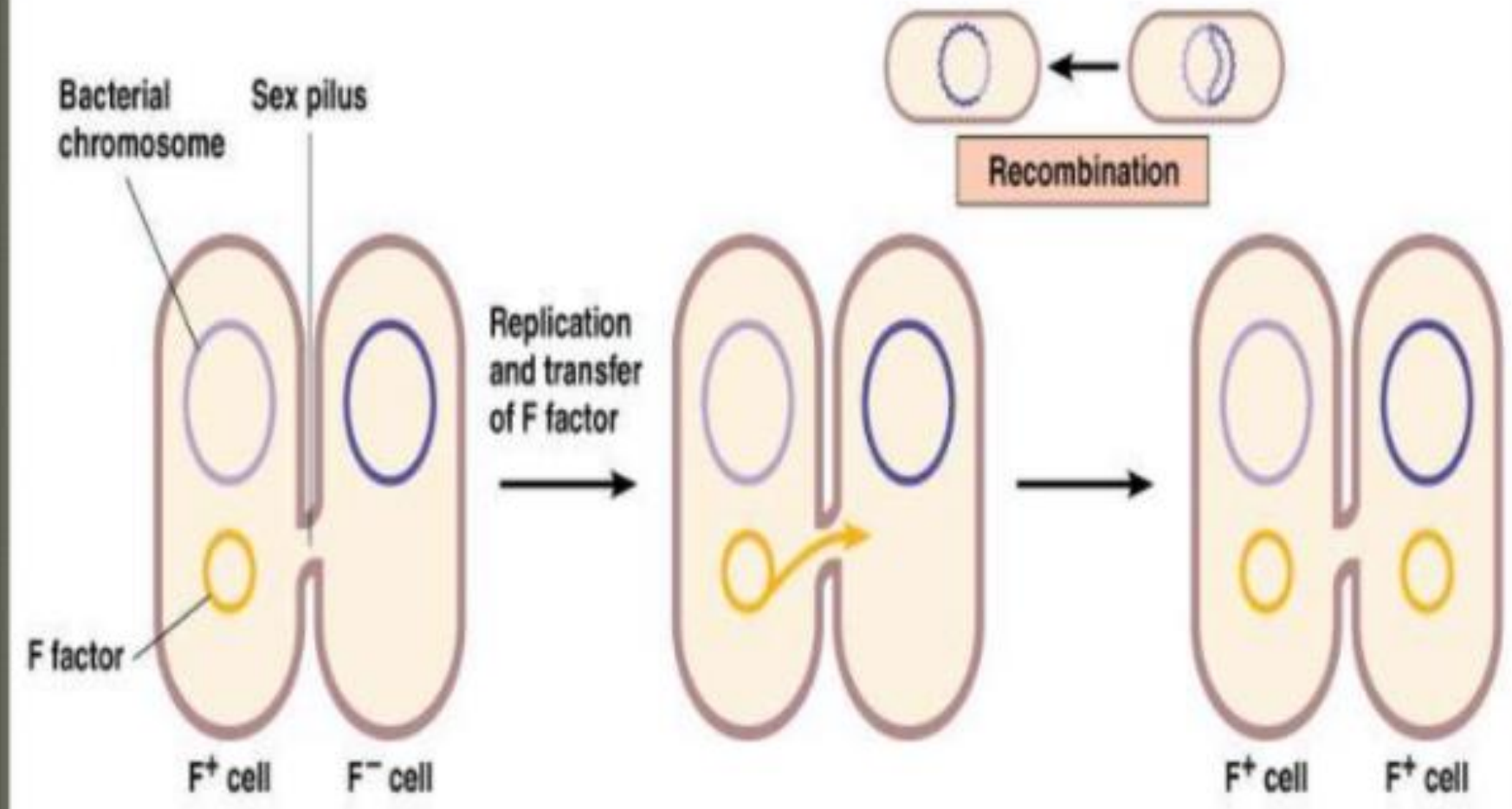
THERE ARE THREE TYPES OF HORIZONTAL GENE TRANSFER

- 1. conjugation: direct transfer of DNA from one bacterial cell to another.
- 2. transduction: use of a bacteriophage (bacterial virus) to transfer DNA between cells.
- 3. transformation: naked DNA is taken up from the environment by bacterial cells.

Conjugation

- Conjugation is the closest analogue in bacteria to eukaryotic sex.
- The ability to conjugate is conferred by the F plasmid.
  - A plasmid is a small circle of DNA that replicates independently of the chromosome.
  - Bacterial cells that contain an F plasmid are called "F+". Bacteria that don't have an F plasmid are called "F-".
- F+ cells grow special tubes called "sex pilli" from their bodies. When an F+ cell bumps into an F- cell, the sex pilli hold them together, and a copy of the F plasmid is transferred from the F+ to the F-. Now both cells are F+.

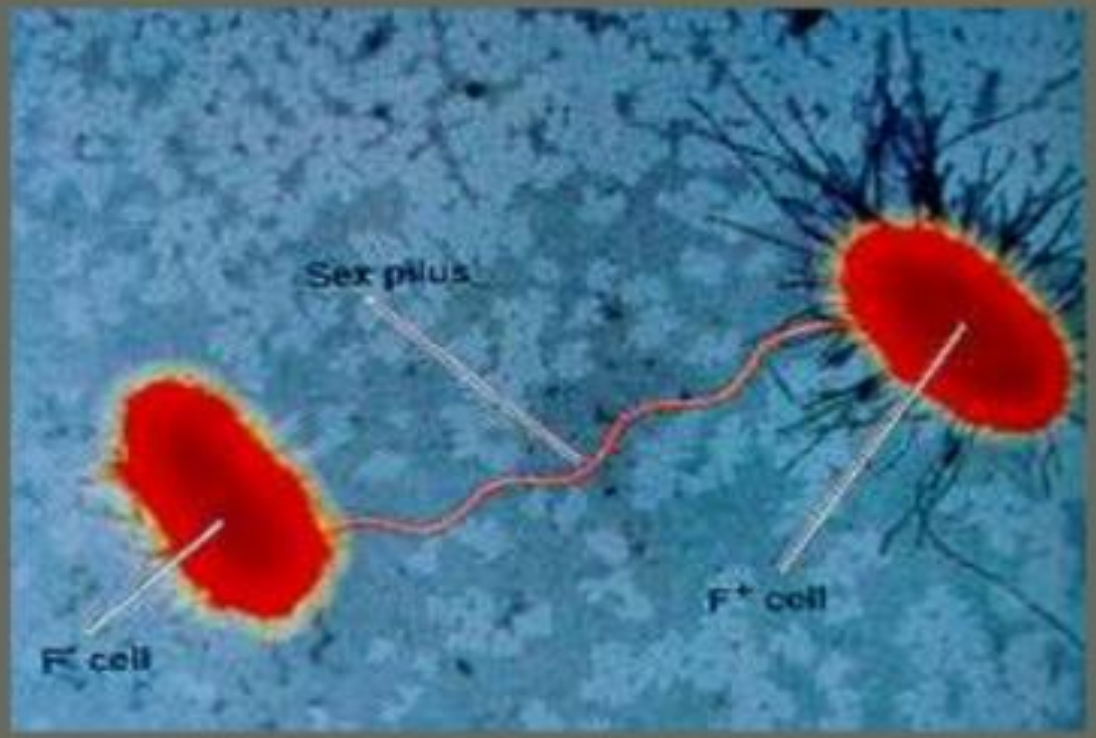
CONJUGATION



(a) When an F factor (a plasmid) is transferred from a donor (F^+) to a recipient (F^-), the F^- cell is converted into an F^+ cell.

Conjugation

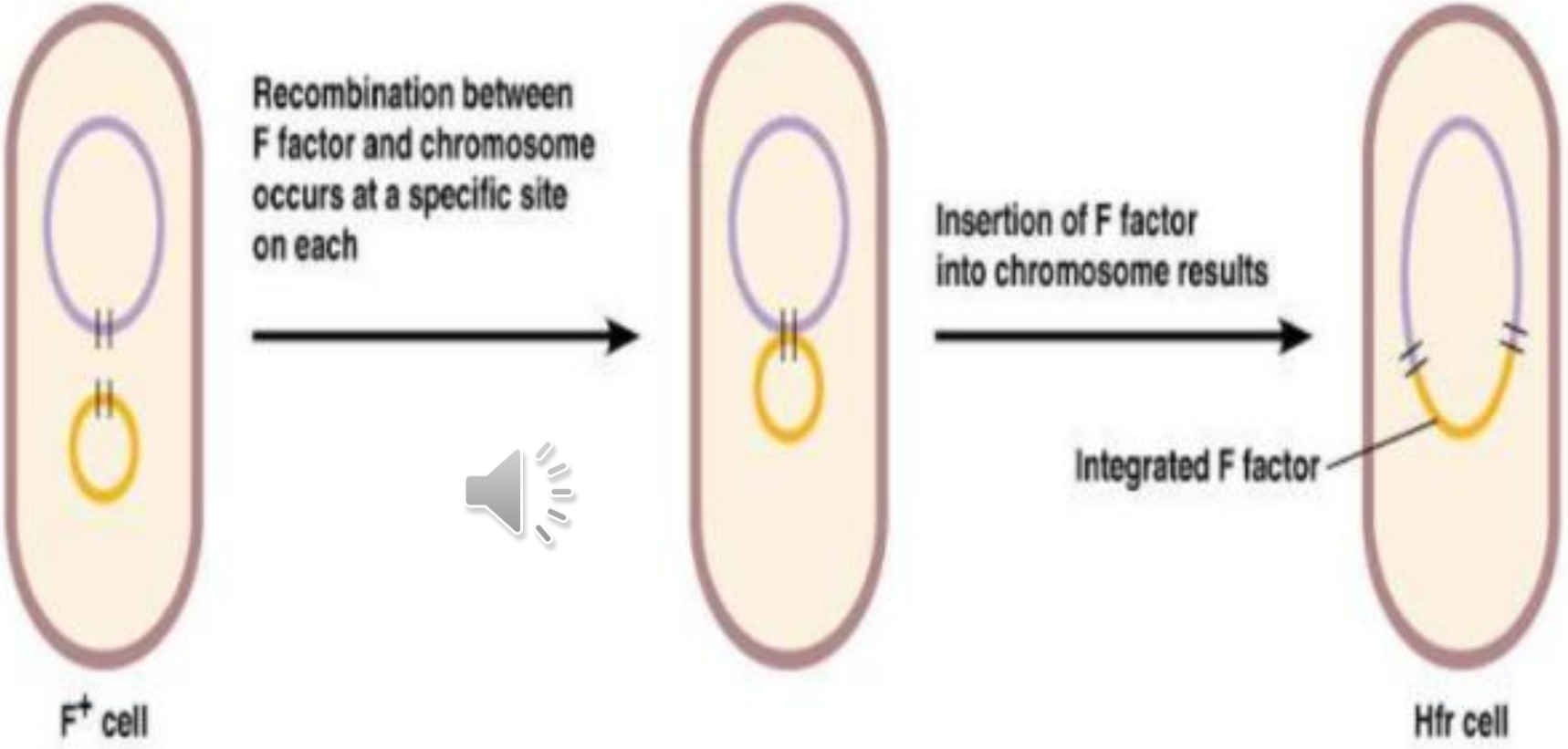
One bacterium passes some DNA (in a plasmid) to another bacterium



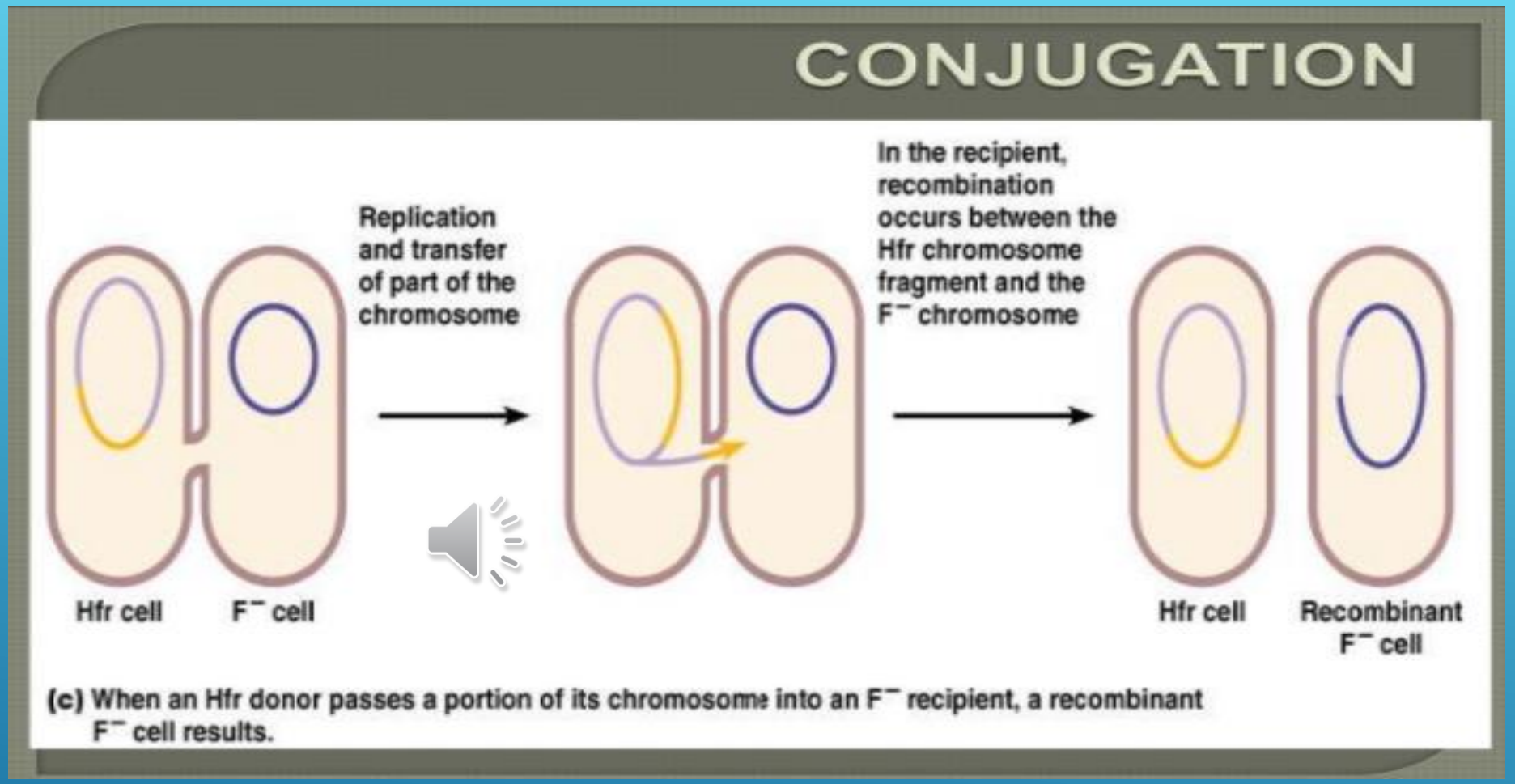
Hfr Conjugation

- sometimes the F plasmid can become incorporated into the bacterial chromosome, by a crossover between the F plasmid and the chromosome.
- The resulting bacterial cell is called an "Hfr", which stands for "High frequency of recombination".
- Hfr bacteria conjugate just like F+ do, but they drag a copy of the entire chromosome into the F- cell.

CONJUGATION



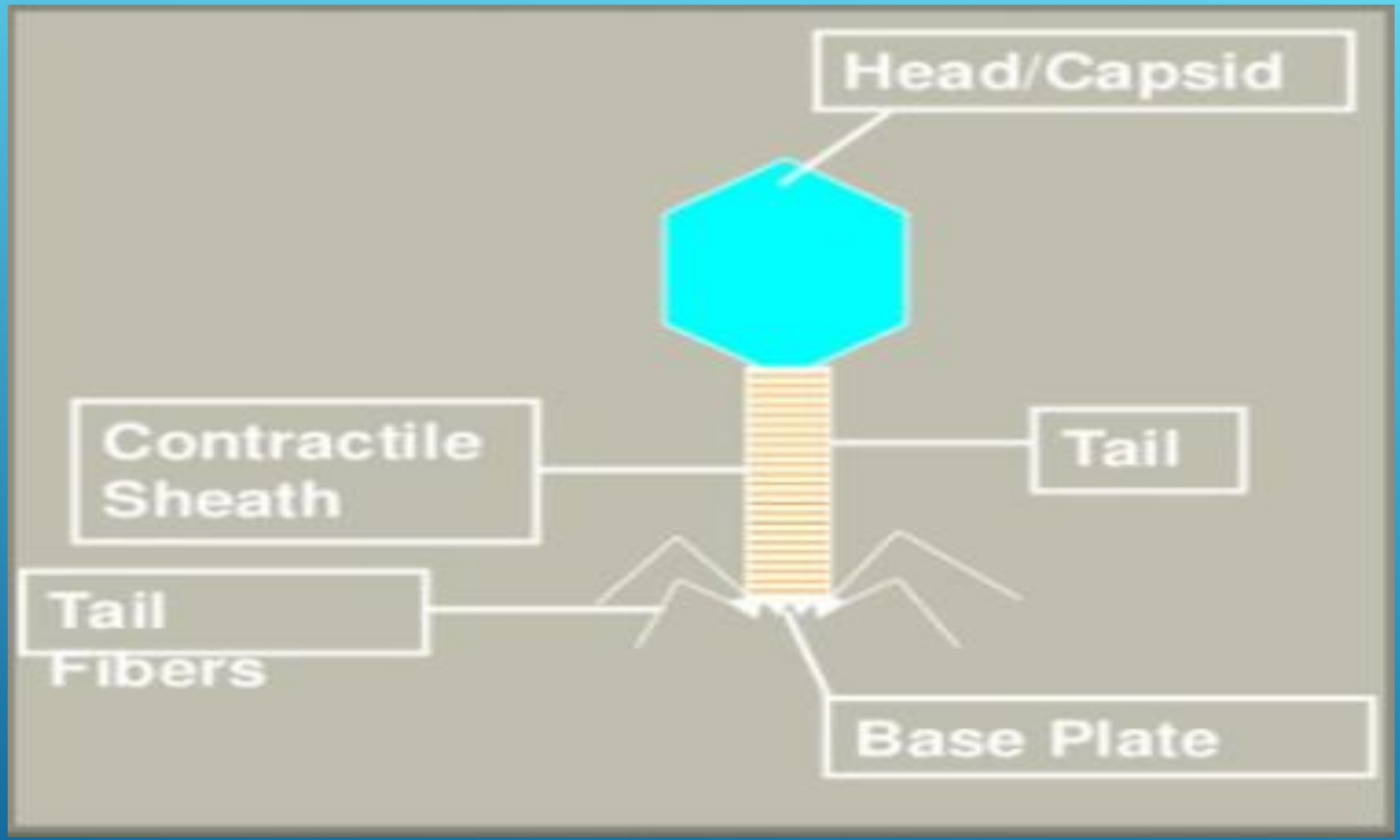
(b) When an F factor becomes integrated into the chromosome of an F^+ cell, it makes the cell a high frequency of recombination (Hfr) cell.



- Hfr bacteria are still able to initiate conjugation with F- cells, but the outcome is completely different from conjugation involving F+ bacteria. IT IS called an F’ instead (F prime)

Transduction

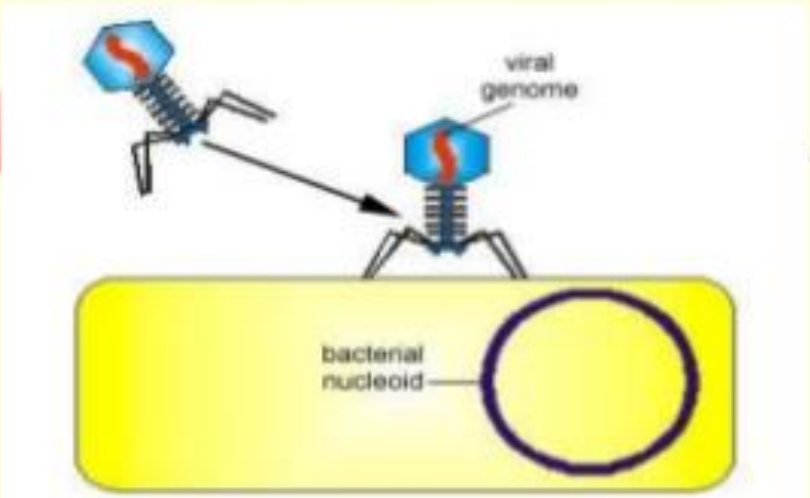
- Transduction is the process of moving bacterial DNA from one cell to another using a bacteriophage.
- Bacteriophage or just “phage” are bacterial viruses.
- Two forms of transduction:
  - 1. generalized: any piece of the bacterial genome can be transferred
  - 2. specialized: only specific pieces of the chromosome can be transferred.


Head/Capsid
Contractile Sheath
Tail
Tail Fibers
Base Plate

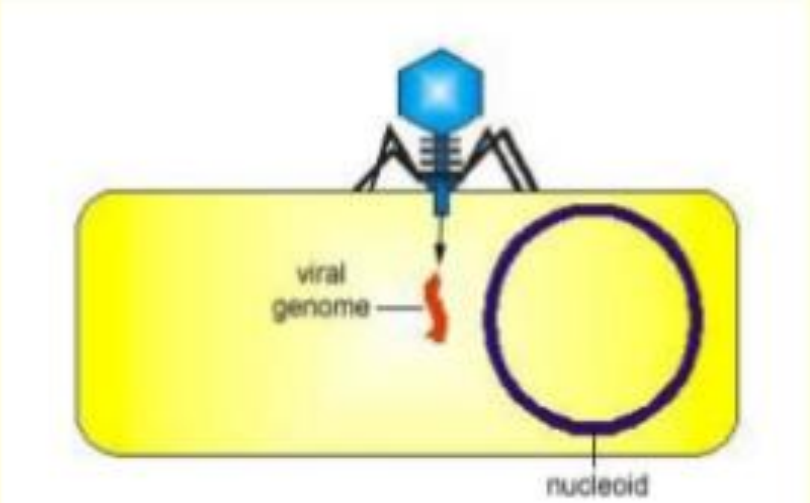
GENERALIZED TRANSDUCTION

- process by which any bacterial gene may be transferred to another bacterium via a bacteriophage.
- typically carries only bacterial DNA and no viral DNA

Generalised Transduction

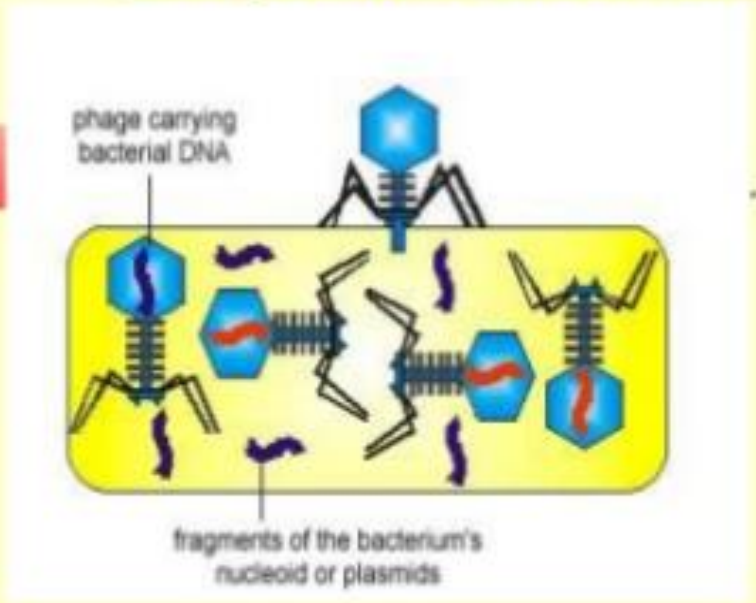


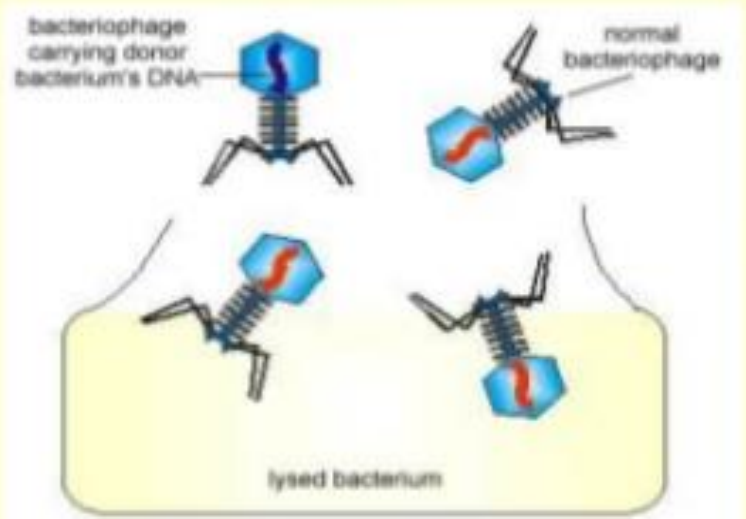
1. A lytic bacteriophage adsorbs to a susceptible bacterium.



2. The bacteriophage genome enters the bacterium. The genome directs the bacterium's metabolic machinery to manufacture bacteriophage components and enzymes

steps in Generalised Transduction (cont'd)



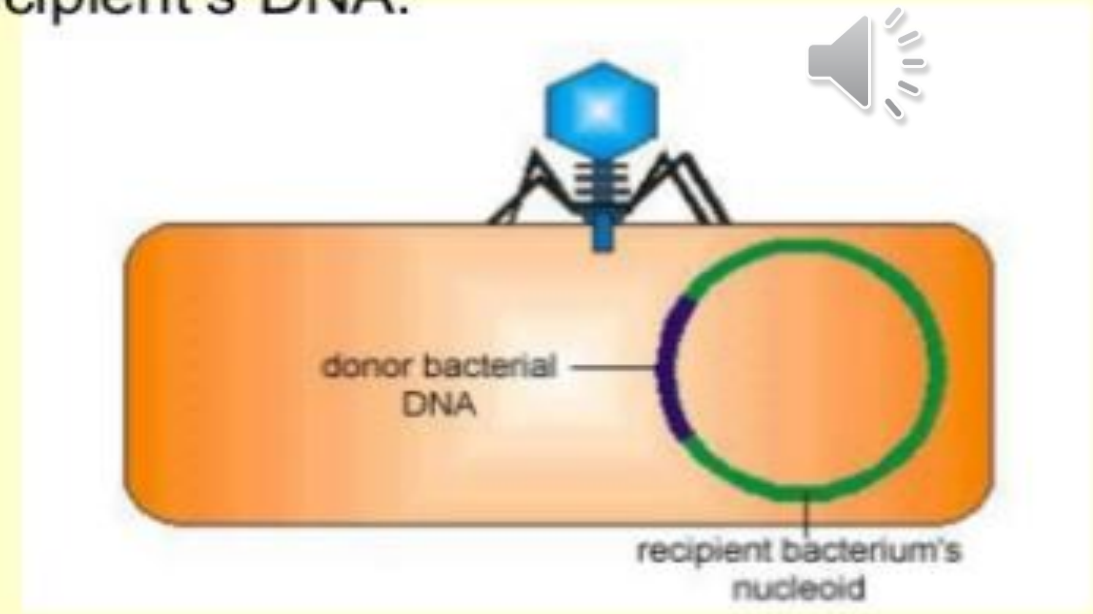


3. Occasionally, a bacteriophage head or capsid assembles around a fragment of donor bacterium's nucleoid instead of a phage genome by mistake.

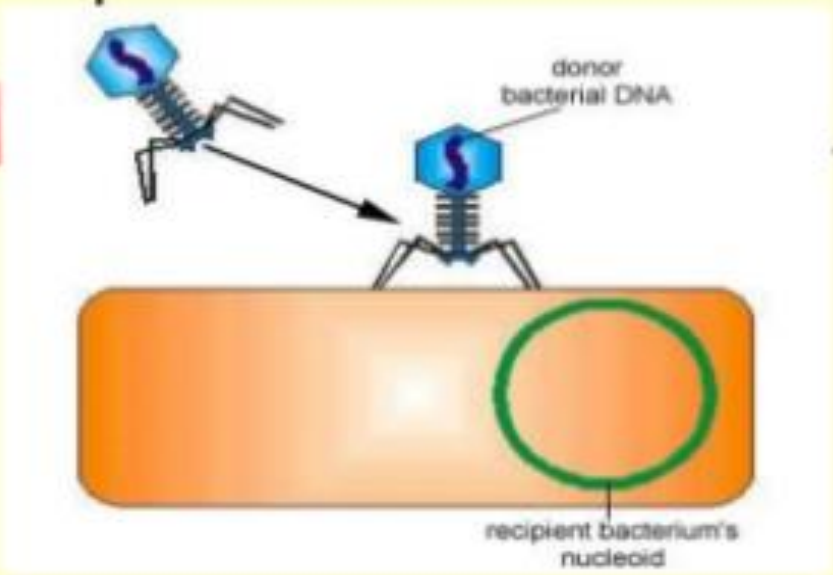
4. The bacteriophages are released

steps in Generalised Transduction (contd)

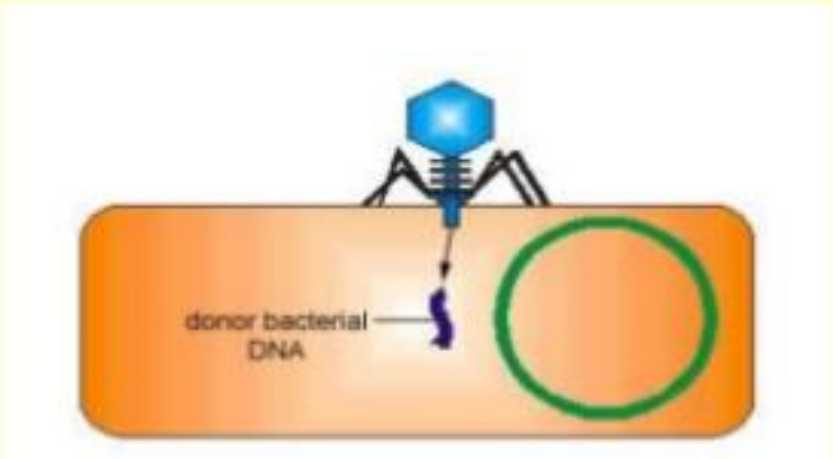
7. The donor bacterium's DNA is exchanged for some of the recipient's DNA.



steps in Generalised Transduction (cont'd)



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 .

Specialized transduction:

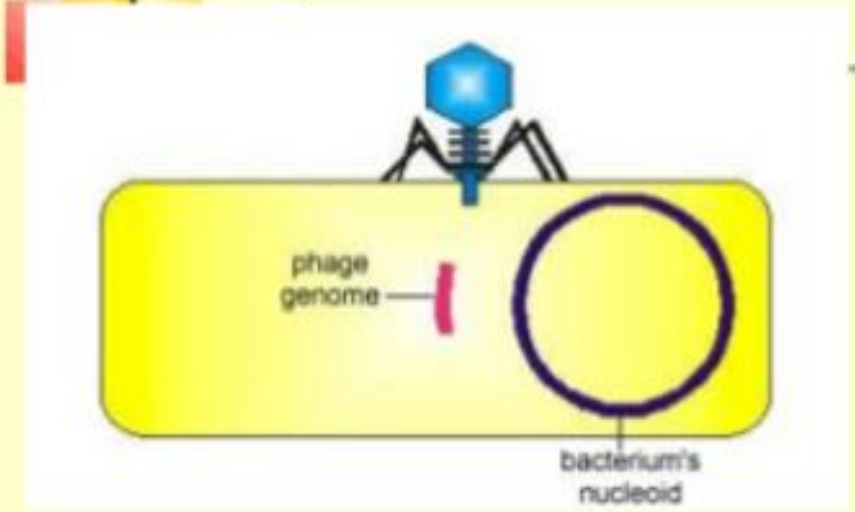
In specialized transduction, bacteriophage transfer only a few restricted gene (DNA fragments) from donor bacteria to recipient bacteria. Specialized transduction is carried only by **temperate bacteriophage .

bacteriophage (phage) are viruses of bacteria - can be either **lytic** or **temperate (Lysogenic)**

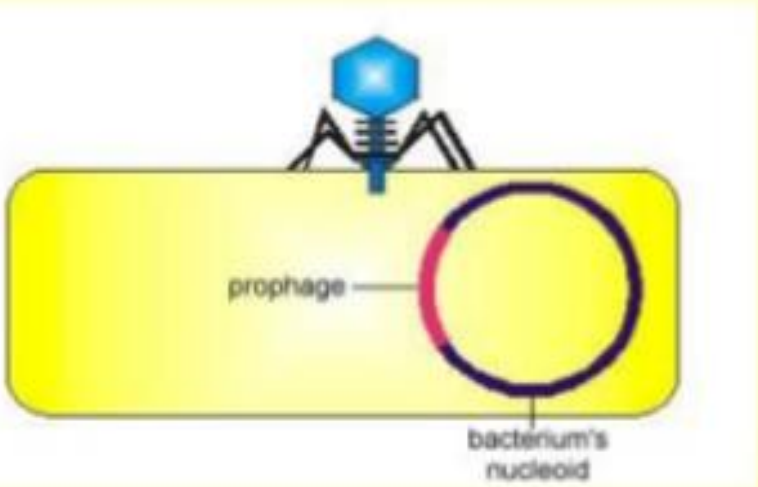
* **lytic** - always lyse (kill) host bacterial cell
* **temperate** - can stably infect and coexist within bacterial cell (**lysogeny**) until a **lytic phase** is induced

* the phage genome during lysogeny is called the **prophage**, and the bacterial cell is called a **lysogen**

Specialised Transduction

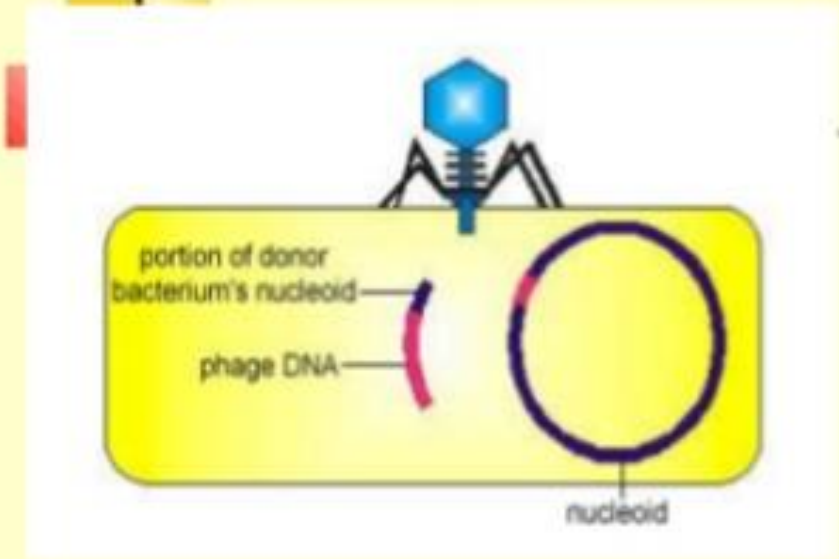


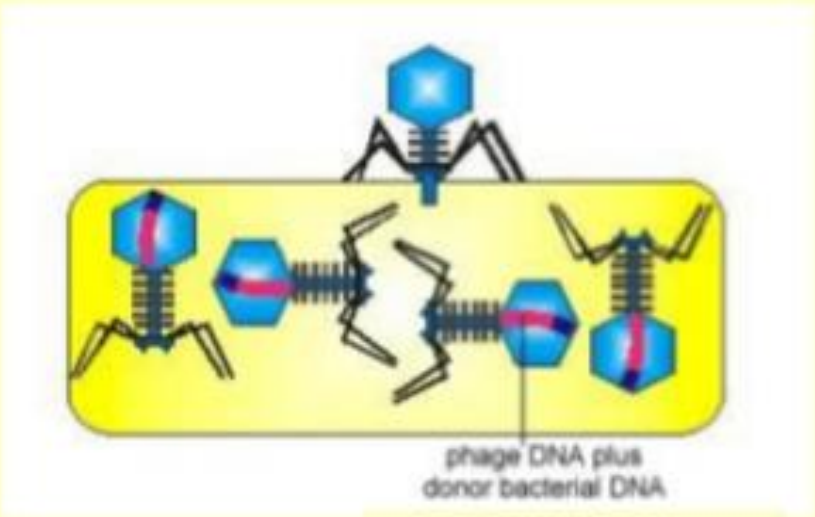
1. A temperate bacteriophage adsorbs to a susceptible bacterium and injects its genome .



2. The bacteriophage inserts its genome into the bacterium's nucleoid to become a prophage.

steps in Specialised Transduction (cont'd)

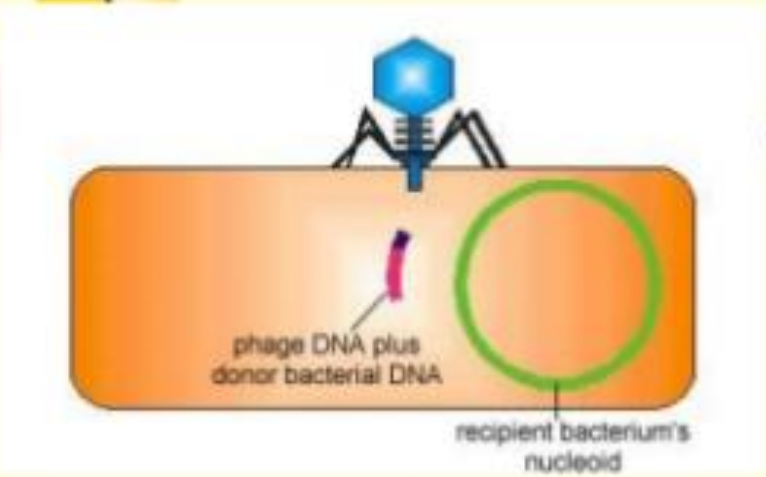




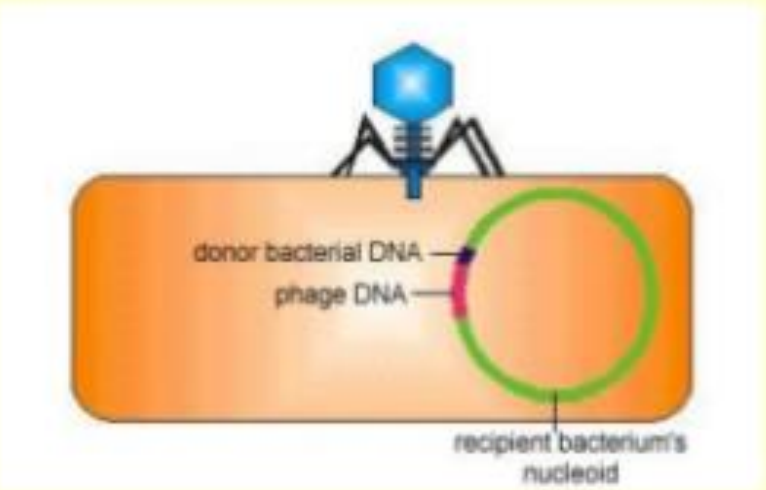
3. Occasionally during spontaneous induction, a small piece of the donor bacterium's DNA is picked up as part of the phage's genome in place of some of the phage DNA which remains in the bacterium's nucleoid.

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.

steps in Specialised Transduction (cont'd)



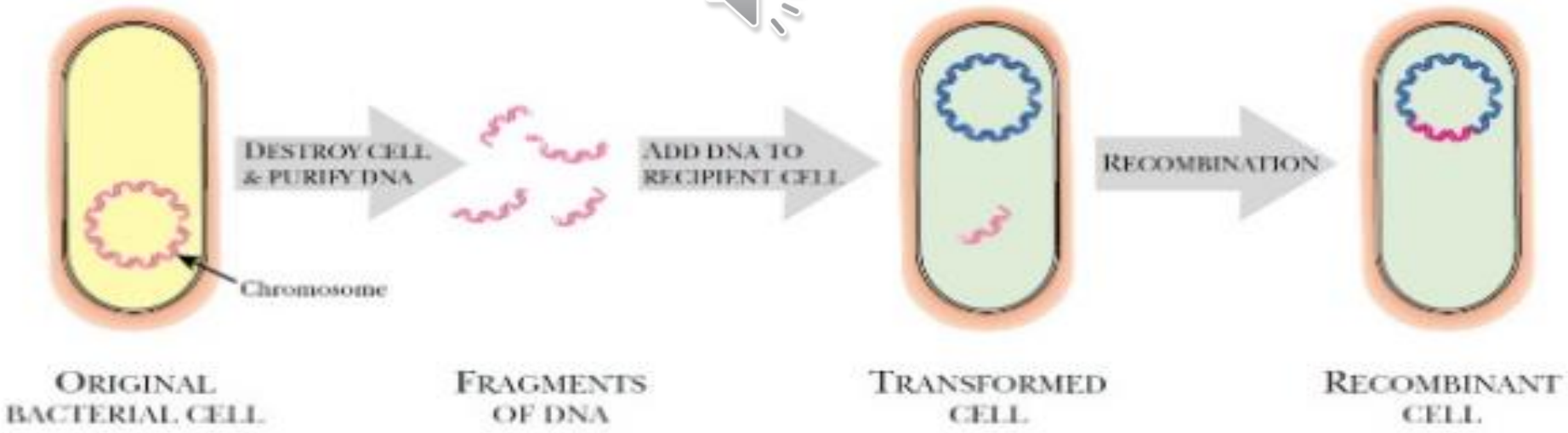
5. The bacteriophage adsorbs to a recipient bacterium and injects its genome.



6. The bacteriophage genome carrying the donor bacterial DNA inserts into the recipient bacterium's nucleoid.

Transformation

The simplest way to transfer genetic information is for one cell to release DNA intothe medium and for another cell to import it.The transfer of “pure” or “naked” DNA from one cell to another is known as transformation.



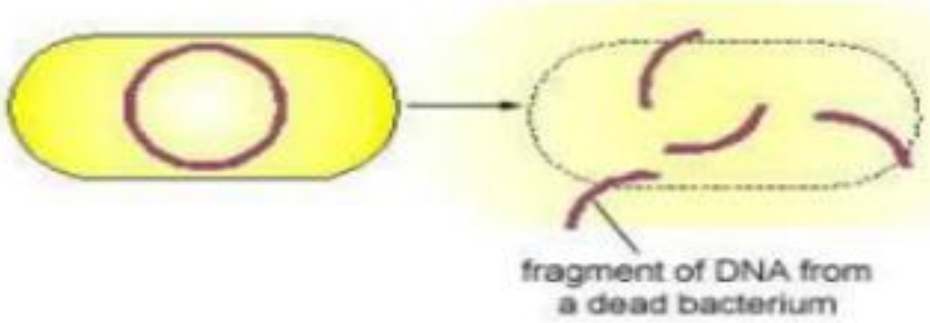
Transformation

By “naked” means that no other biological macromolecules, such as protein, are present to enclose or protect the DNA.

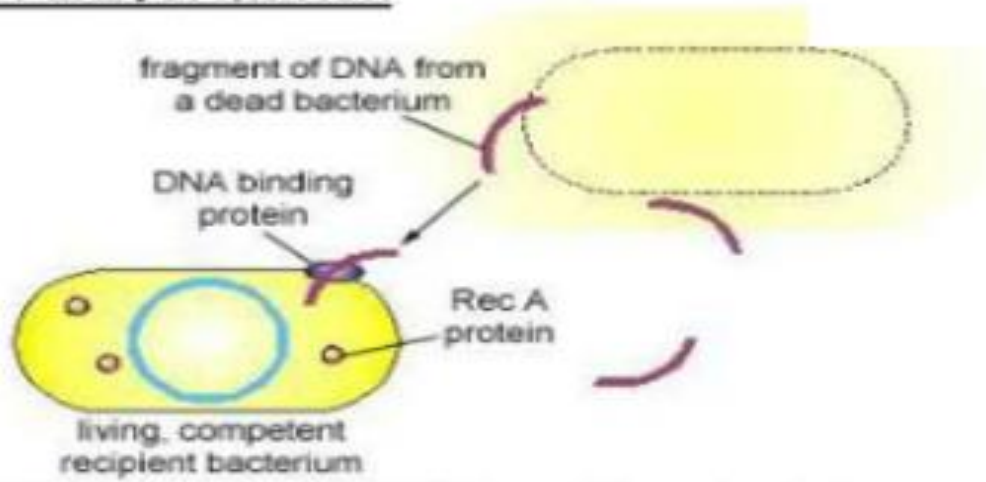
No actual cell-to-cell contact is involved in transformation, nor is the DNA packaged inside a virus particle.

In practice, transformation is mostly a laboratory technique.The DNA is extracted
from one organism by the experimenter and offered to other cells in culture. Cells able
to take up DNA are said to be “**competent**.”

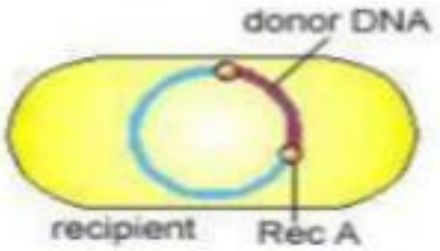
The 4 steps in Transformation



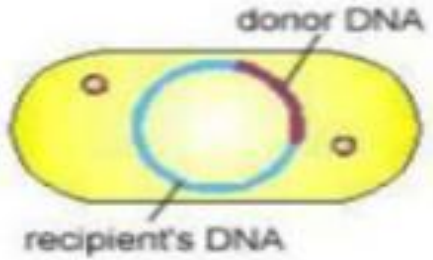
1. A donor bacterium dies and is degraded



2. A fragment of DNA from the dead donor bacterium binds to DNA binding proteins on the cell wall of a competent, living recipient bacterium



3. The Rec A protein promotes genetic exchange between a fragment of the donor's DNA and the recipient's DNA



4. Exchange is complete