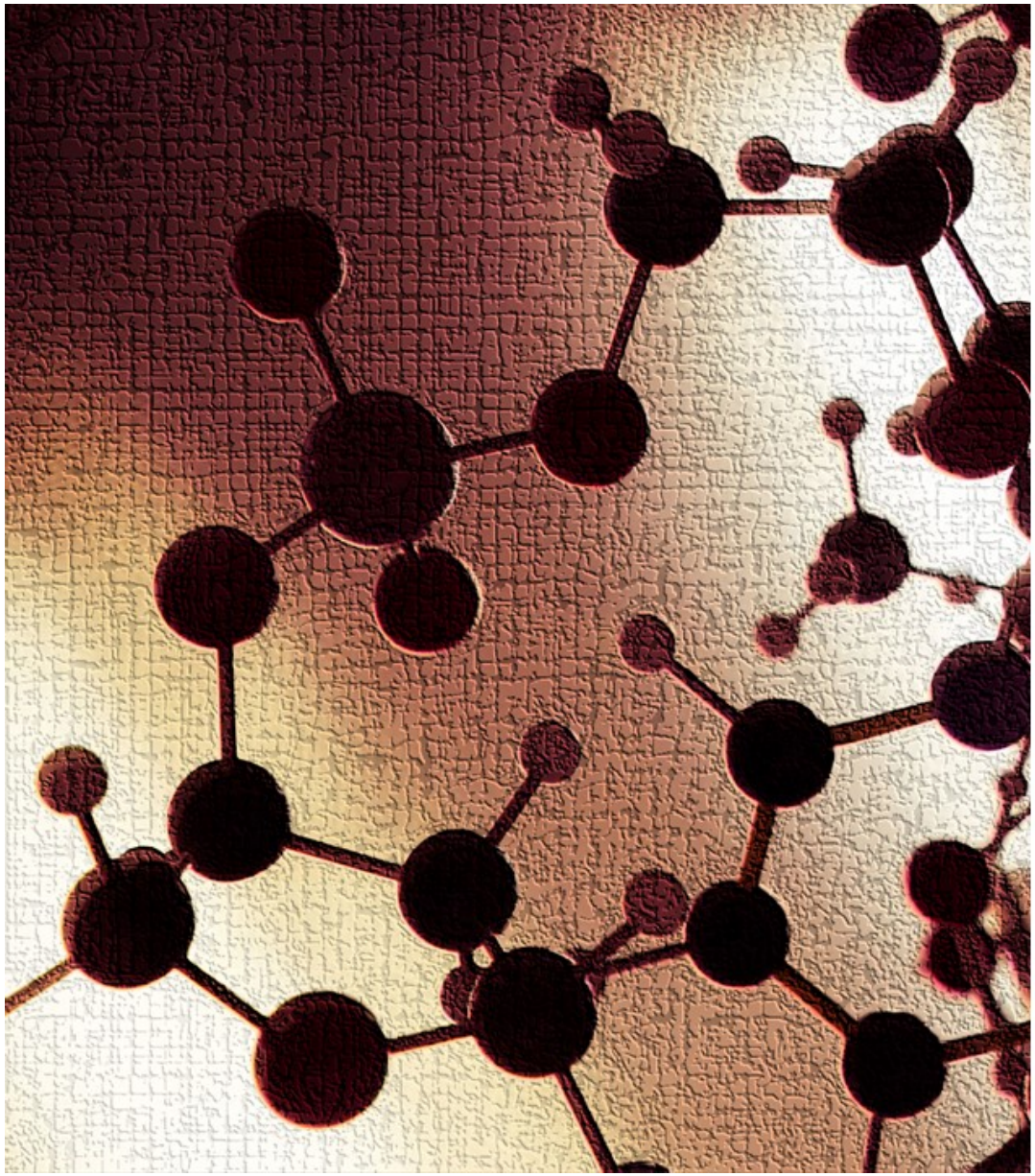

Undergraduate course, Monsoon semester, 2020

2nd Professional year (Veterinary Microbiology)

Topic: Bacterial genetics

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Genotypic variations :

- **Genotypic variation occurs due to**
  - Mutation
  - Acquisition or loss of gene

Mutations: Chemical Basis and Effects

- Stable, heritable changes in sequence of bases in DNA that lead to phenotypic
- point mutations
  - from substitution of single pairs of nucleotide
  - from the addition or deletion of nucleotide pairs
- larger mutations
  - insertions, deletions, inversions, duplication, and translocations of nucleotide sequences

Cont…

- Mutation are of two types based on inducing agent
  i. Spontaneous mutation: arises naturally
  ii. Induced mutation: occurs as effect of mutagen
- Effect of mutation
  - Mutations may be deleterious
  - Mutations may be beneficial
  - Mutations effect may have no effect
  - mutant may survives and becomes an allele

Acquisition of new genes

- Horizontal gene transfer (HGT) allows introduction of genetic material from one organism to another within the same generation
- It is an important way to introduce genetic diversity.
- HGT even permit distantly related species to share genes, influencing their phenotypes.
- The HGT and mutation are significant sources of genetic variation, the raw material for the process of natural selection, in prokaryotes.

Types of mutation

- **Transversion mutations** – substitution of purine and pyrimidine bases
- **Missense Mutation**
- **Nonsense Mutation**
- **Silent Mutation**
- **Frameshift mutations**
  - Insertion
  - Deletion

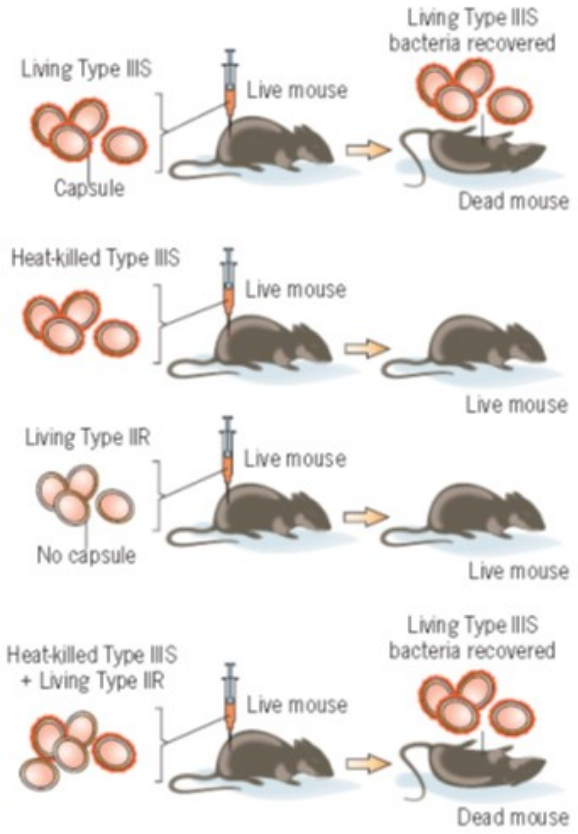
Transformation
(L. *trans,* across, and *formatio,* to form).

- Transformation is **uptake of naked DNA molecules** from surrounding and subsequent **integration into the bacterial chromosome** by homologous recombination

- The bacterial transformation first observed by Fred Griffith (1928) while studying pneumococcal infection in mice, (*Streptococcus pneumoniae*)

Bacterial transformation

- **Figure:** Griffith's discovery of transformation in *Streptococcus pneumoniae*



Significance of Transformation

- Transformation enables bacterial populations to
  - overcome great fluctuations in population dynamics
  - mitigate the challenge of maintaining the population numbers during harsh and extreme environmental changes.
  - adapt for survival and evolution

Purpose of DNA uptake

- Following purposes of DNA uptake by bacteria:
    - DNA for genetic diversity
    - DNA repair
    - DNA as nutritional need

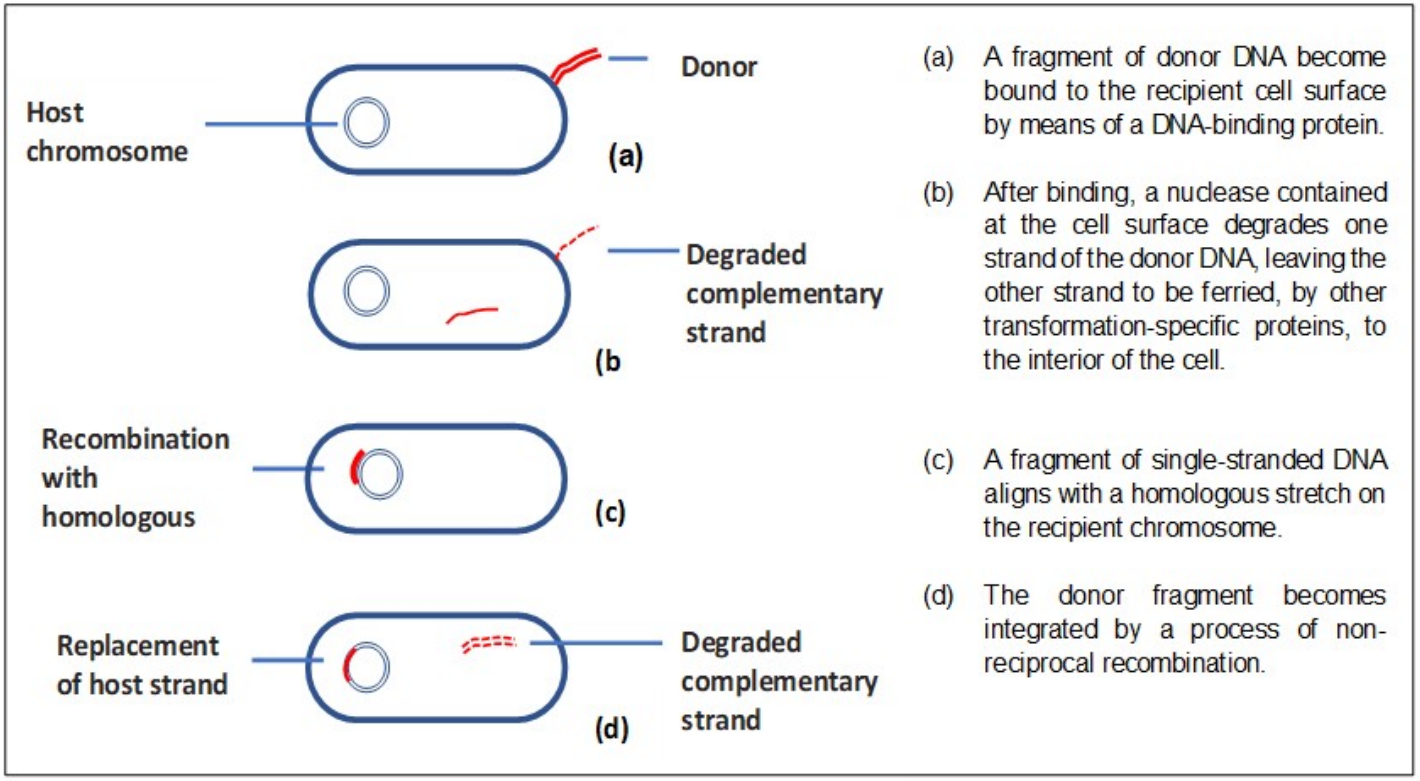
Competence for transformation

- **Competence** - Specialized physiological state of bacterial cell due to which uptake of exogenous DNA takes place.
- Competence generally occurs in late logarithmic phase
- The induction of competence occurs by two process:
    a) Natural competence
    b) Induced competence

Cont…

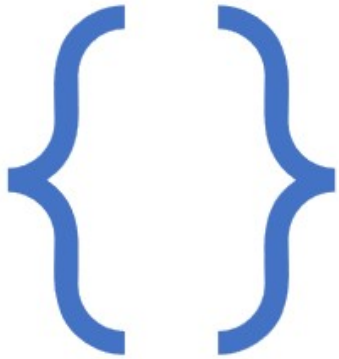
- **Natural competence**
  - genetically programmed physiological state in which prokaryotes are able to take up genetic material from their surroundings.
- **Induced competence**
  - laboratory procedures that involve making the cell passively permeable to DNA
- The recipient bacteria are made artificially competent by two methods:
  i. Chemical treatment
  ii. Electroporation

Figure: Transformation in bacteria



Applications of Transformation

- Transformation can be used as host cells for the following:
  - DNA linkage studies and gene mapping
  - For making multiple copies of the DNA
  - For cloning procedures in laboratory
  - To express large amounts of proteins and enzymes
  - For the generation of cDNA libraries



Transduction (L. *transducere*, to lead across).

- The process of transferring genetic material from one bacterium to another by a virus is referred as transduction.

- first described by Zinder and Lederberg for Salmonella and phage P22

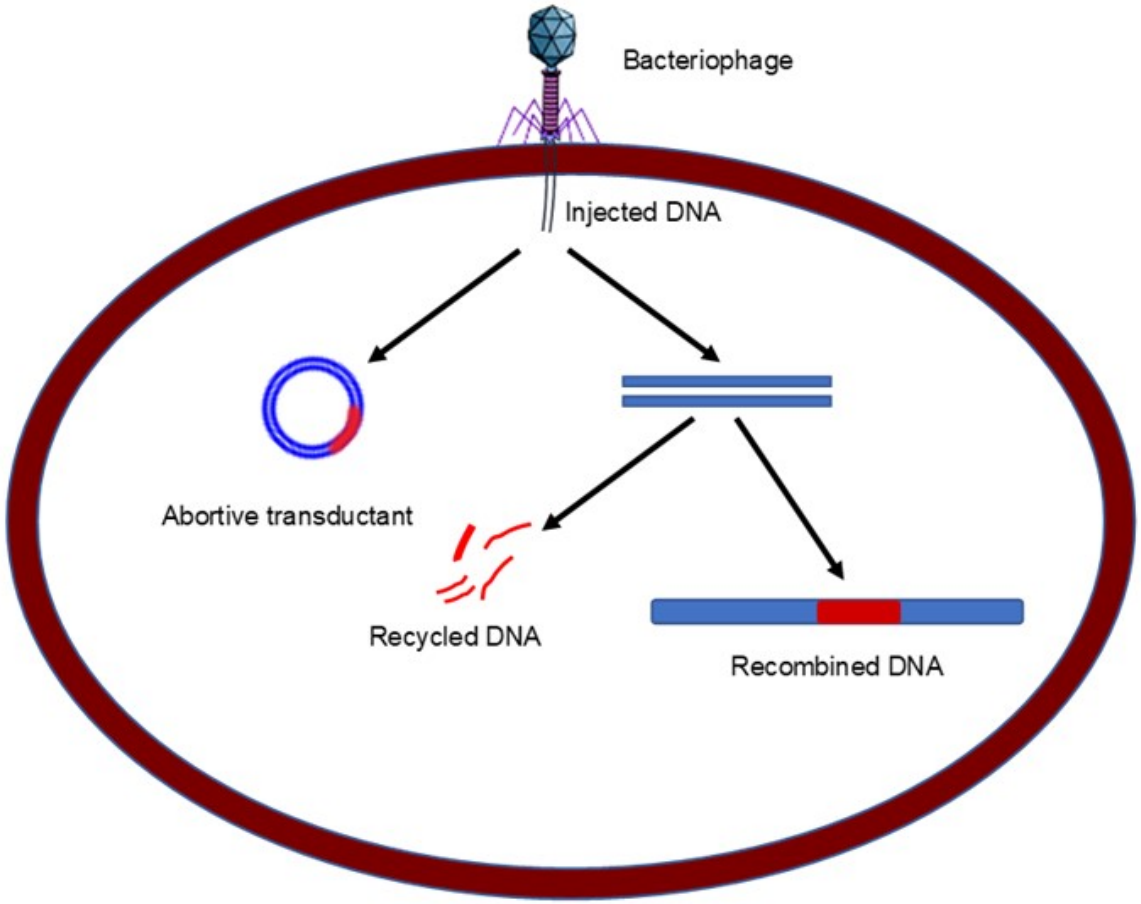


Figure : Fate of transferred DNA in generalized transduction

Formation of transductant

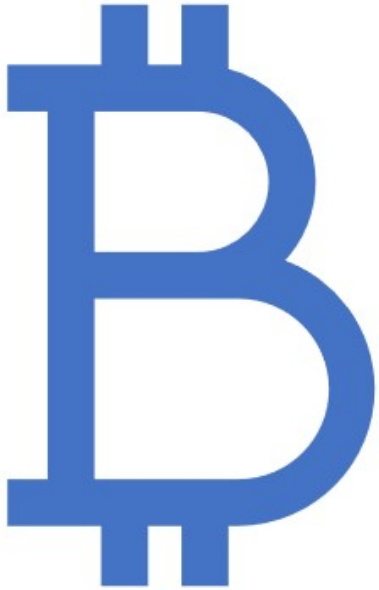
- errors in fidelity during packaging of DNA into the phage heads lytic growth
- bacteriophages occasionally remove a fragments of the host cell's bacterial DNA
- phage-like particles contain a segment of bacterial genome (donor cell).
- virions carrying host DNA may transfer to other bacterial (recipient cell)

Condition for transduction

- phage degrade the bacterial chromosome (lytic cycle);
- process of packaging DNA into the phage protein not be specific for phage DNA;
- bacterial genes transferred by the virus recombine with the chromosome in the recipient cell.

Types of transduction

- Two types
  - Generalised transduction
  - Specialised transduction



There are two different types of transduction.

1. **Generalized transduction:** a random or nearly random fragment of bacterial DNA is packaged in the phage head in place of the phage chromosome.

2. **Specialized transduction:** a recombination event occurs between the host chromosome and the phage chromosome, producing a phage chromosome that contains a piece of bacterial DNA.

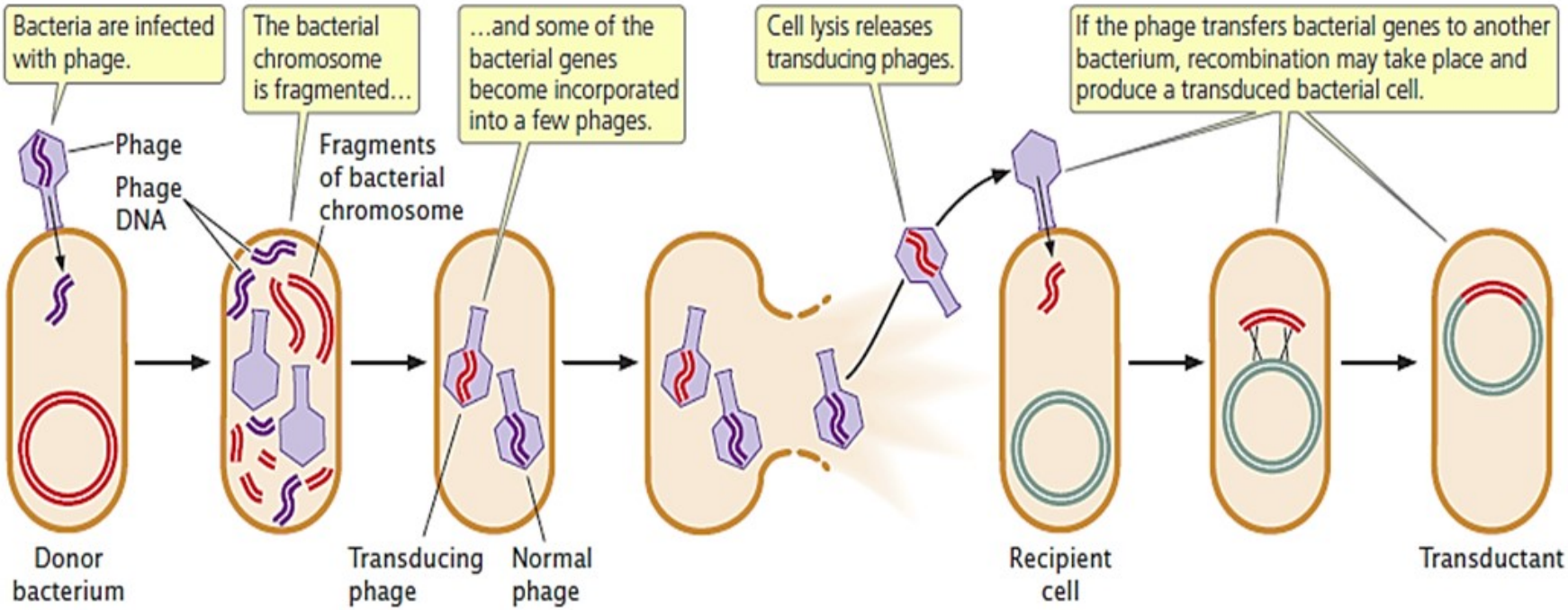
- Phage particles that contain bacterial DNA are called *transducing particles. Generalized transducing particles* contain only bacterial DNA. *Specialized transducing particles* always contain both phage and bacterial DNA.

Generalized Transduction

- Generalized transducing phages can transport any bacterial gene from one cell to another—thus, the name generalized transduction.

- The best known generalized transducing phages are P22 in *S. typhimurium* and *P1 in E. coli.*

Generalized transduction

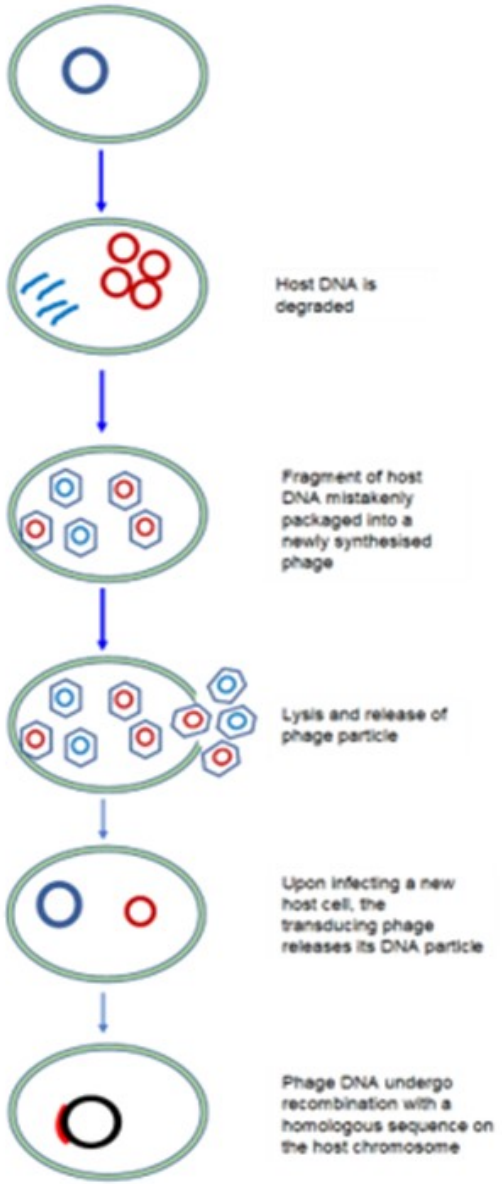


Specialized Transduction

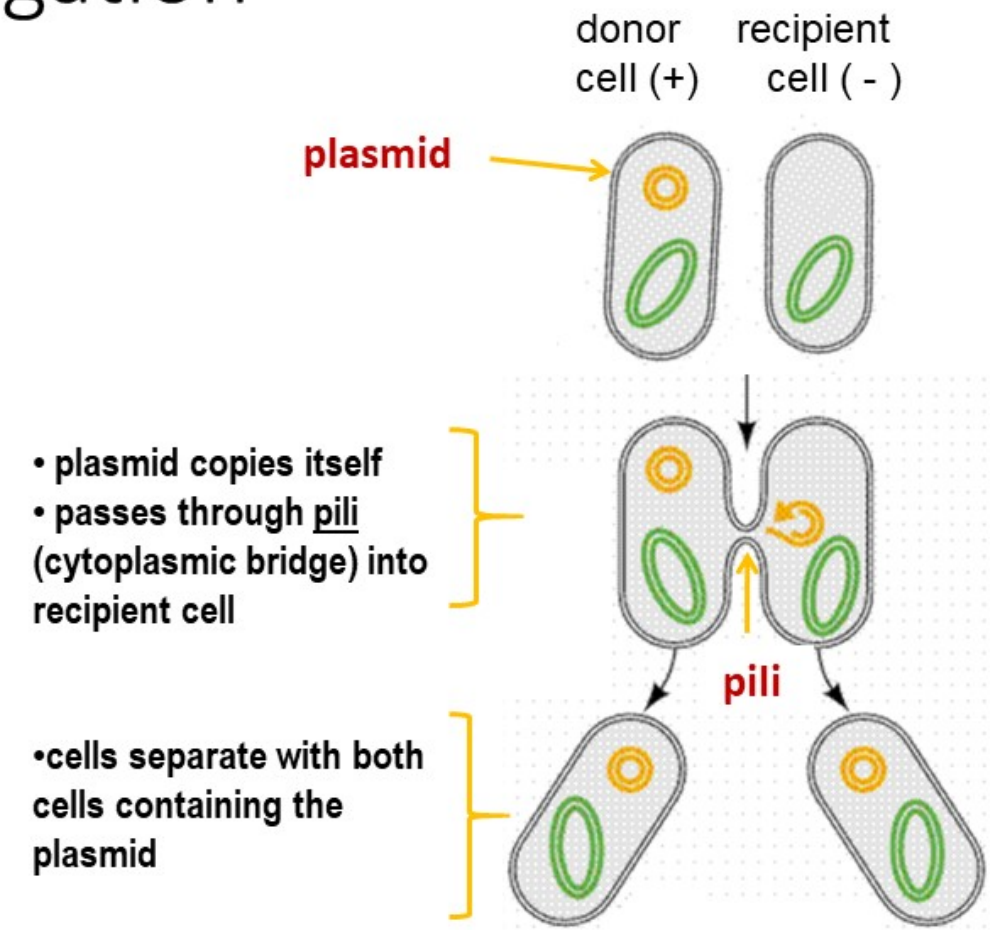
- Specialized transduction is characteristic of viruses that transfer only certain genes between bacteria.
- Bacteriophage lambda is the best-known specialized transducing phage; carries only the *gal* (required for the utilization of galactose as an energy source) and *bio* (essential for the synthesis of biotin) *genes from one E. coli* cell to another. The insertion site is between the *gal genes and the bio* on the *E. coli chromosome.*
- The integrated chromosome — the prophage—in a lysogenic cell undergoes rare (about one in 105 cell divisions) spontaneous excision, whereupon it enters the lytic pathway.
- Prophage excision can also be induced, for example, by irradiating lysogenic cells with ultraviolet light. Normal excision is essentially the reverse of the site-specific integration process and yields intact circular phage and bacterial chromosomes.

Steps of specialised transduction

- temperate bacteriophage adsorbs to a susceptible bacterium and injects its genome .
- The bacteriophage inserts its genome carrying part of the donor DNA into the recipient
- The donor DNA integrates with recipient DNA
- The prophage replicates along with host DNA and inherited to daughter cell



Bacterial Conjugation

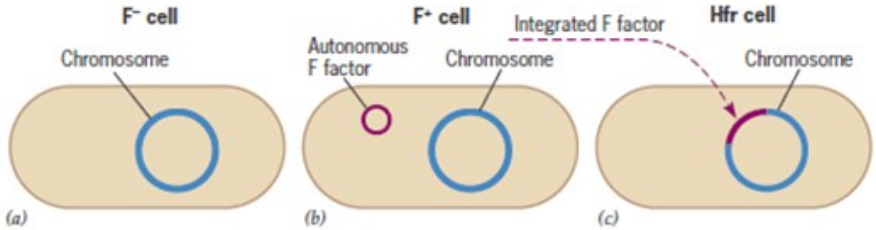


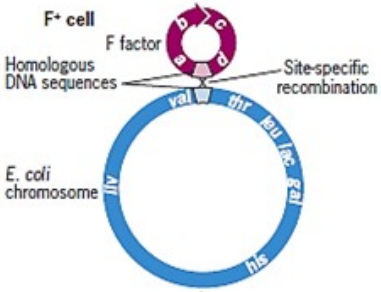
- In 1946, Joshua Lederberg and Edward Tatum discovered that *E. coli cells transfer genes by conjugation.*
- Conjugation has proven to be an important method of genetic mapping in bacterial species.
- The donor (F+) and recipient cells (F-) are in direct contact during conjugation.
- Types:

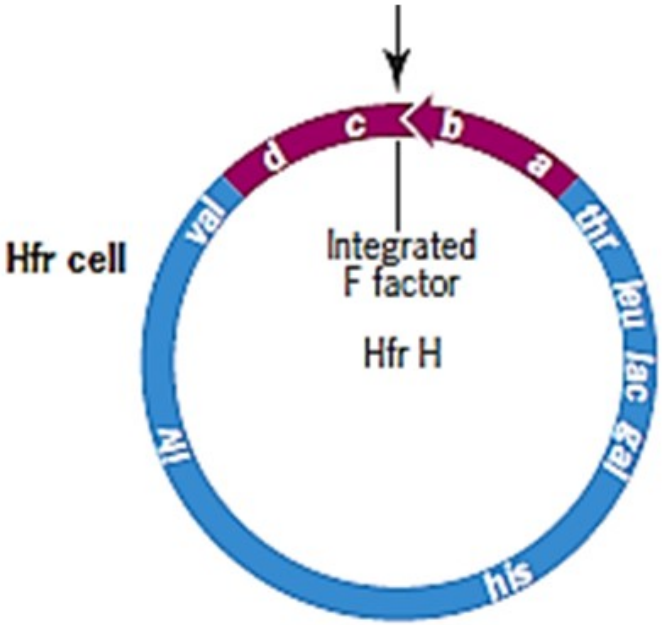
  1. F+ conjugation

  2. Resistance plasmid conjugation

  3. Hfr (high frequency recombinant) conjugation


F⁻ cell
Chromosome
F⁺ cell
Autonomous
F factor
Chromosome
Integrated F factor
Hfr cell
Chromosome
(a)
(b)
(c)


F⁺ cell
F factor
Homologous
DNA sequences
Site-specific
recombination
E. coli
chromosome


Hfr cell
Integrated
F factor
Hfr H

F' FACTORS AND SEXDUCTION

- An Hfr strain is produced by the integration of an F factor into the chromosome by recombination between IS elements in the chromosome and IS elements in the F factor.
- Indeed, rare F+ cells are present in Hfr cultures, indicating that excision of the F factor does occur (by a process that is essentially the reverse of the integration event Moreover, anomalous excision events produce autonomous F factors carrying bacterial genes.
- These modified F factors, called F' ("F prime") factors, were first identified by Edward Adelberg and Sarah Burns in 1959.
- Transfer of F' factors to recipient (F-) cells is called **sexduction;** it occurs by the same mechanism as F factor transfer in F+ X F -mating carrying up to half the bacterial chromosome.


Any questions????

Thank you