Mechanisms of Infectious Disease • Fall 2006

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Genetic Basis of Variation in Bacteria

I. Organization of genetic material in bacteria
- a. chromosomes
- b. plasmids

II. Genetic variation: Source
- a. point mutations
- b. DNA rearrangements

III. Genetic variation: Transmission
- a. transformation
- b. transduction
- c. conjugation

IV. Genetic variation: Implications for pathogenesis

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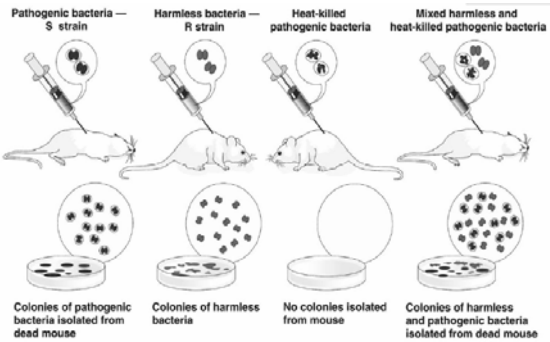
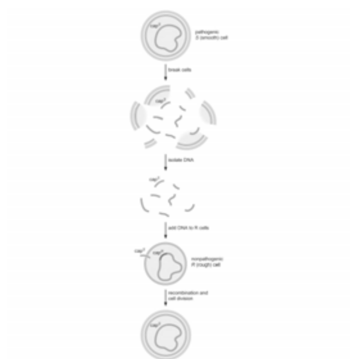
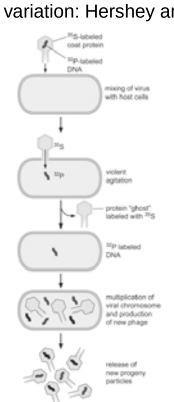
Genetic basis of variation: Griffiths (1928)



Genetic basis of variation: Avery et.al (1944)



Genetic basis of variation: Hershey and Chase (1952)



Organization of genetic material in bacteria: chromosomes

- **Most** bacteria contain a single chromosome (+ extrachromosomal elements)
- **Some** bacteria have been found also to contain 2-3 replicons which can be considered either megaplasmids or minichromosomes e.g. 3.0 Mb and 0.9 Mb replicons in *Rhodobacter sphaeroides*
- A **few** bacterial genera contain >1 chromosome e.g. 2.1 Mb and 1.2 Mb chromosomes in *Brucella*
- **Some** bacteria harbour large replicons essential for survival in a specific ecological niche but not under laboratory conditions e.g. 1.4 Mb and 1.7 Mb replicons in*Rhizobium meliloti* are required for plant symbiosis
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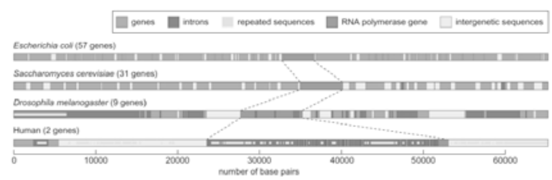
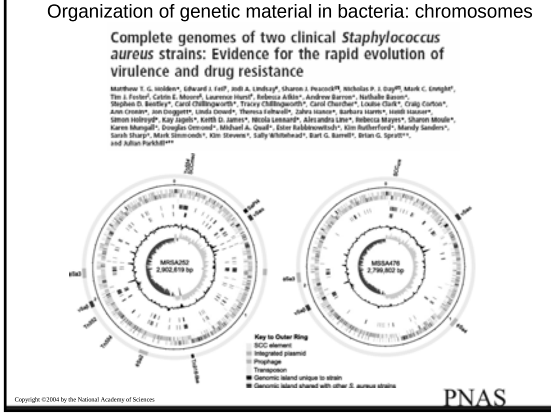
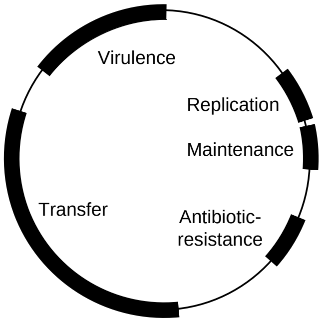
Organization of genetic material in bacteria: chromosomes



Organization of genetic material in bacteria: chromosomes



Organization of genetic material in bacteria: plasmids



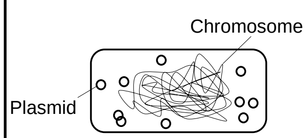
Organization of genetic material in bacteria: plasmids

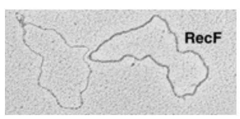
Examples of naturally-occuring plasmids and relevant features

Plasmid	Host	Plasmid size (kb)	Relevant feature
pT181	*Staphylococcus aureus*	4.4	Tetracycline resistance
ColE1	*Escherichia coli*	6.6	Colicin production and immunity
pGKL2	*Kluyveromyces lactis*[b]	13.5	Killer plasmid
pAMβ1	*Enterococcus faecalis*	26.0	Erythromycin resistance
pSK41	*Staphylococcus aureus*	46.4	Multidrug resistance
pBM4000	*Bacillus megaterium*	53.0	rRNA operon
pI258	*Staphylococcus aureus*	28.0	Metal ion resistance
pSLT	*Salmonella enterica* subsp. *typhimurium*	93.9	Virulence determinants
pMT1	*Yersinia pestis*	101.0	Virulence determinants
pADP-1	*Pseudomonas* sp.	108.8	Atrazine (herbicide) catabolism
pWW0	*Pseudomonas putida*	117.0	Aromatic hydrocarbon degradation
pBtoxis	*Bacillus thuringiensis* subsp. *israelensis*	137.0	Mosquito larval toxicity
pX01	*Bacillus anthracis*	181.7	Exotoxin production
pSOL1	*Clostridium acetobutylicum*	192.0	Solvent production
pSymB	*Sinorhizobium meliloti*	1683.3	Functions associated with plant symbiosis

Organization of genetic material in bacteria: plasmids

- Extrachromosomal
- Circular or linear
- 2 kb to hundreds of kb in size
- Non-essential
- May carry 'supplemental' genetic information or may be cryptic
- Employ host functions for most of DNA metabolism





http://www.biochem.wisc.edu/inman/empics/dna-prot.htm

Sources of genetic variation: point mutations

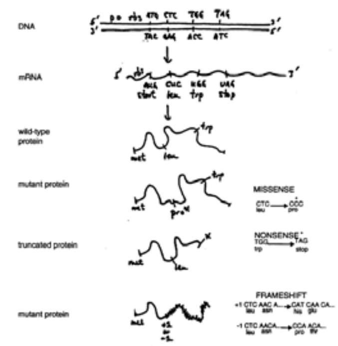
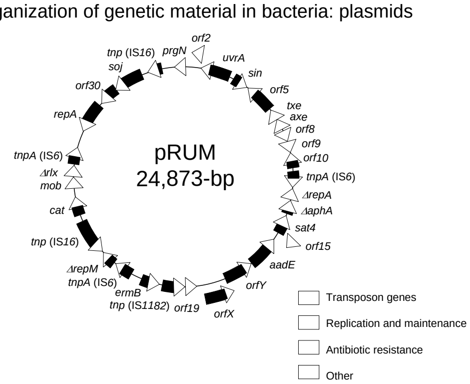
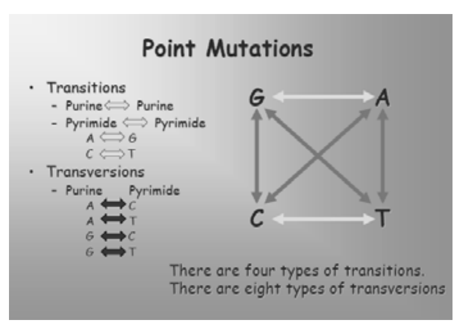


Fig. 3. Point mutations.

Organization of genetic material in bacteria: plasmids



Sources of genetic variation: point mutations



Sources of genetic variation: point mutations

Mutation phenotypes

- Silent mutation (synonymous), no change in amino acid
  AGG > AGA , both codons specify Arginine
- Missense mutation (replacement; nonsynonymous), change in amino acid
  - Nonsynonymous missense (or radical replacement)
    UUU (Phe) > UCU (Ser); Phe is hydrophobic and Ser is polar
- Nonsense mutation, premature termination of translation
  CAG (Gln) > UAG (Stop)
- Frameshift, addition or deletion of base pairs, not in a multiple of three, within the coding region of a gene.

Sources of genetic variation: DNA rearrangements

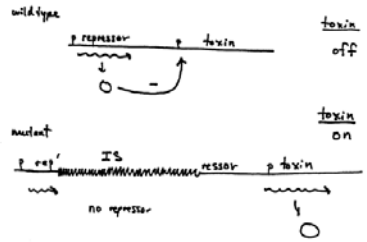
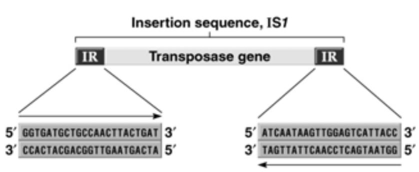


Fig. 5 (at left). Disruption of a gene by IS element transposi-

Sources of genetic variation: DNA rearrangements

<u>Insertion sequence (IS) elements</u>:

1. **Simplest type of transposable element found in bacterial chromosomes and plasmids.**
2. **Encode only genes for mobilization and insertion.**
3. **Range in size from 768 bp to 5 kb.**
4. **<u>IS1</u> first identified in *E. coli*'s glactose operon is 768 bp long and is present with 4-19 copies in the *E. coli* chromosome.**
5. **Ends of all known IS elements show <u>inverted terminal repeats</u> (<u>ITRs</u>).**



Sources of genetic variation: DNA rearrangements

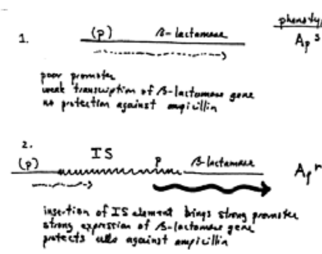
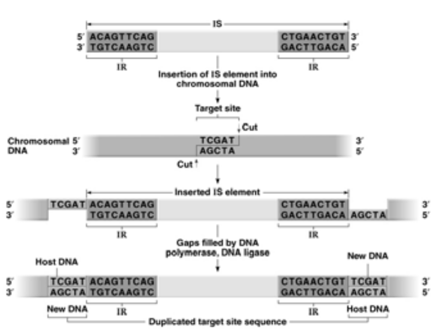


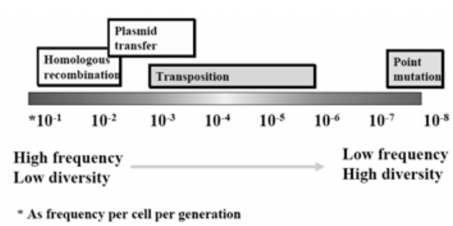
Fig. 6 (at left). Activation of a gene by IS element transposition.

Sources of genetic variation: DNA rearrangements

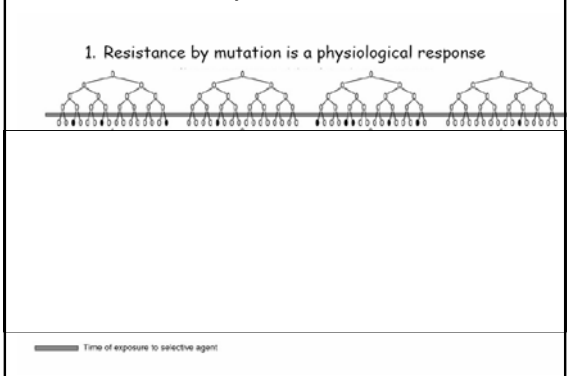
Integration of IS element in chromosomal DNA.



Sources of genetic variation: frequency of occurrence



Transmission of genetic variation: Luria-Delbruck test



Linear transmission of genetic variation

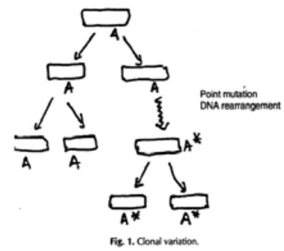
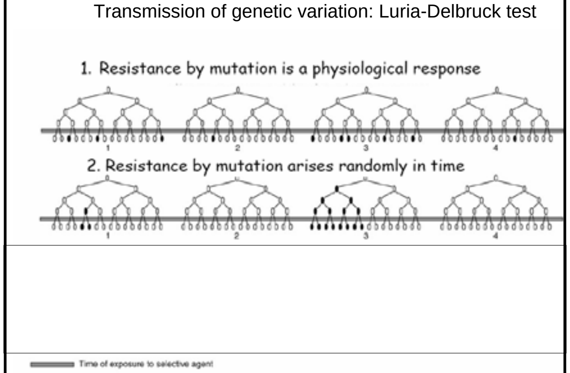


Fig. 1. Clonal variation.

Transmission of genetic variation: Luria-Delbruck test



Horizontal transmission of genetic variation

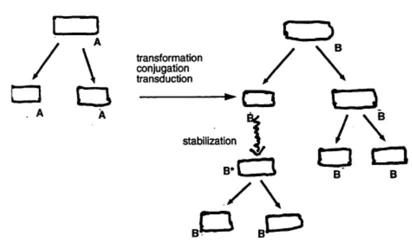
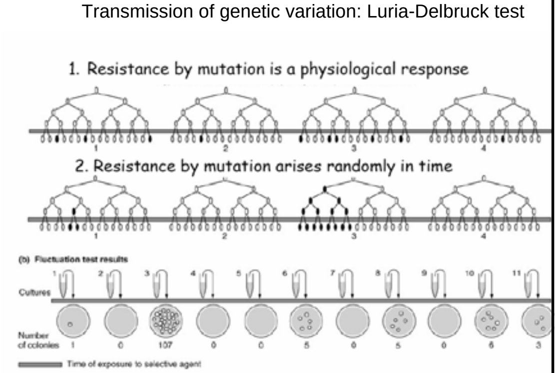


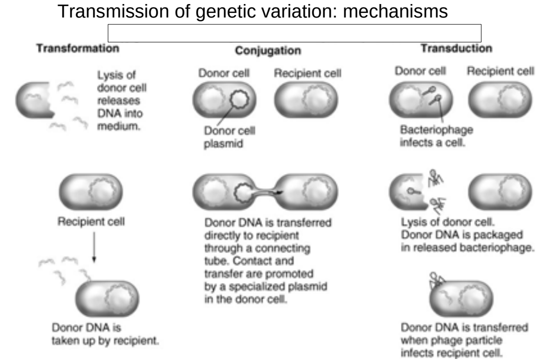
Fig. 2 *(at left).* Variation by transfer of genetic information.

Transmission of genetic variation: Luria-Delbruck test



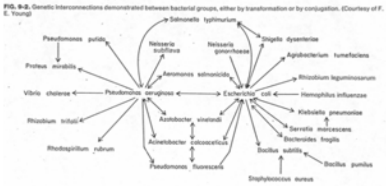
Results fit with expectations if random mutation occur at random.

Transmission of genetic variation: mechanisms



Transmission of genetic variation: mechanisms

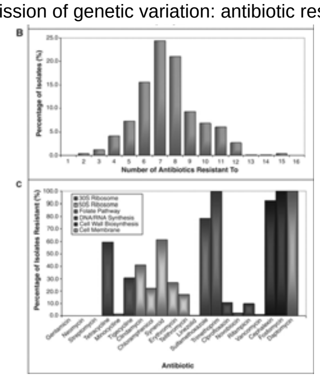
FIG. 9-2. Genetic interconnections demonstrated between bacterial groups, either by transformation or by conjugation. (Courtesy of F. E. Young)



Transmission of genetic variation: transformation

- Gene transfer resulting from the uptake of DNA from a donor.
- Factors affecting transformation
  - DNA size and state
    - Sensitive to nucleases
  - Competence of the recipient (*Bacillus, Haemophilus, Neisseria, Streptococcus*)
    - Competence factor
    - Induced competence

Transmission of genetic variation: antibiotic resistance



V. M. D'Costa et al. Science (2006)

Fig. 1. Antibiotic resistance profiling of 480 soil-derived bacterial isolates

Transmission of genetic variation: transformation

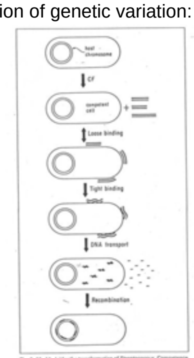
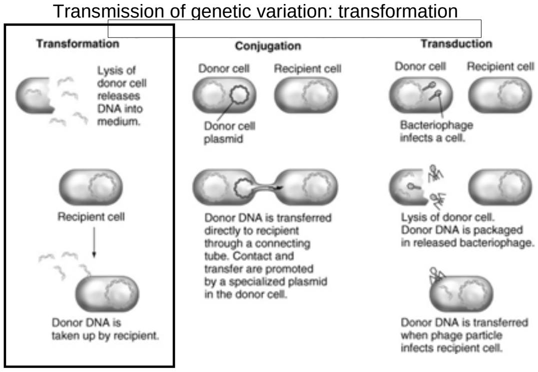
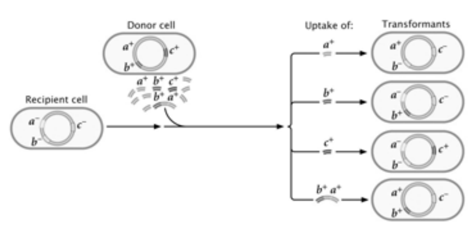


Fig. 6-11. Model for the transformation of Streptococcus. Competence factor (CF) is required for the induction of the competent state. Duplex DNA binds to competent cells in at least two distinct steps, referred to as loose and tight binding. Transport of single-strand fragments results in intracellular protein-DNA complexes, and subsequent recombination of the fragments into the host chromosome.

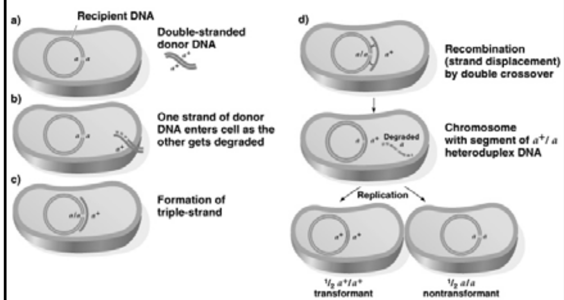
Transmission of genetic variation: transformation



Transmission of genetic variation: transformation



Transmission of genetic variation: transformation

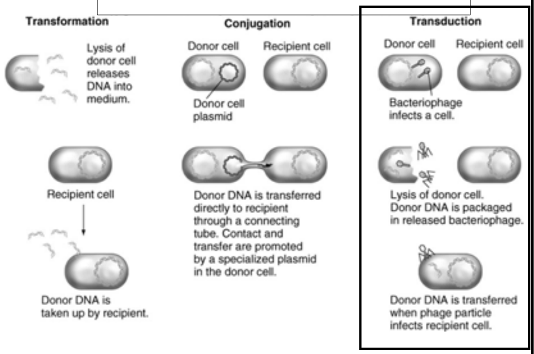


Transmission of genetic variation: transduction

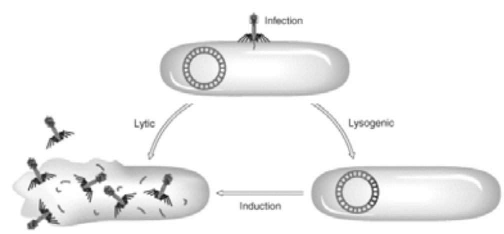
How did Zinder and Lederberg prove that the phenotype was the result of transduction?

- presence of DNAase rules out transformation
- filter prevented contact so no conjugation
- reducing filter pore size to below size of phage inhibited

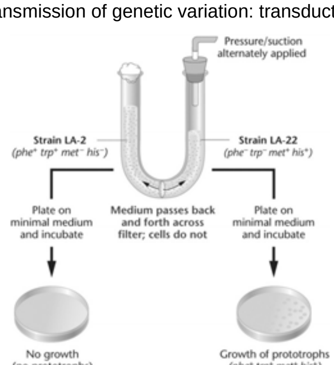
Transmission of genetic variation: transduction



Transmission of genetic variation: transduction



Transmission of genetic variation: transduction

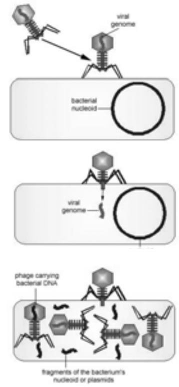


Transmission of genetic variation: transduction

QuickTime™ and a
TIFF (LZW) decompressor
are needed to see this picture.

Transmission of genetic variation: transduction

Transmission of genetic variation: generalized transduction

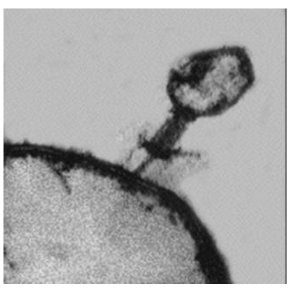


1. A <u>lytic bacteriophage</u> adsorbs to a susceptible bacterium.

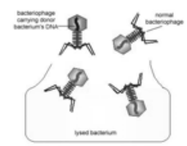
2. The bacteriophage genome enters the bacterium. The genome directs the bacterium's metabolic machinery to manufacture bacteriophage components and enzymes

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.

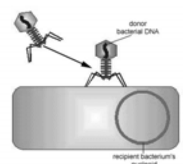
Transmission of genetic variation: transduction



Transmission of genetic variation: generalized transduction



4. The bacteriophages are released.

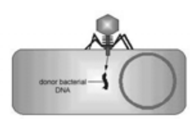


5. The bacteriophage carrying the donor bacterium's DNA adsorbs to a recipient bacterium

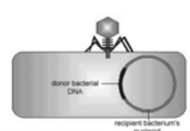
Transmission of genetic variation: transduction

- There are two types of transduction:
  - 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

Transmission of genetic variation: generalized transduction



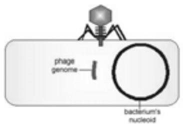
6. The bacteriophage inserts the donor bacterium's DNA it is carrying into the recipient bacterium .



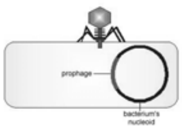
7. The donor bacterium's DNA is exchanged for some of the recipient's DNA.

http://www.cat.cc.md.us/courses/bio141/lecguide/unit4/genetics/recombination/transduction/transduction.html

Transmission of genetic variation: specialized transduction

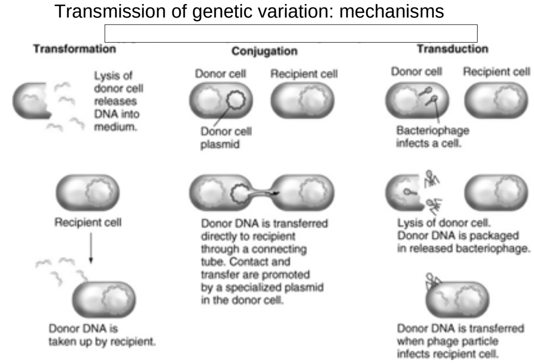


1. A temperate bacteriophage adsorbs to a susceptible bacterium and injects its genome .

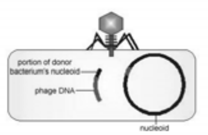


2. The bacteriophage inserts its genome into the bacterium's nucleoid to become a prophage.

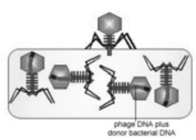
Transmission of genetic variation: mechanisms



Transmission of genetic variation: specialized transduction

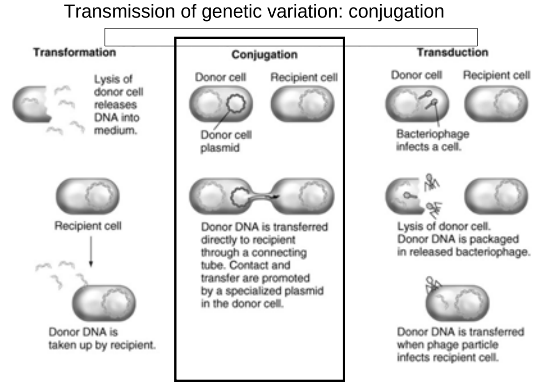


3. Occasionally during spontaneous induction, a small piece of the donor bacterium's DNA is picked up as part of the phage's genome in place of some of the phage DNA which remains in the bacterium's nucleoid.

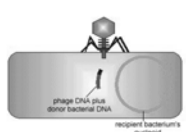


4. As the bacteriophage replicates, the segment of bacterial DNA replicates as part of the phage's genome. Every phage now carries that segment of bacterial DNA.

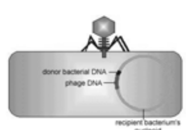
Transmission of genetic variation: conjugation



Transmission of genetic variation: specialized transduction



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.

http://www.cat.cc.md.us/courses/bio141/lecguide/unit4/genetics/recombination/transduction/spectran.html