## Transformation in Bacteria

In transformation, ―naked‖ DNA in solution is transferred from one bacterial cell (donor cell) to another (recipient cell). It can occur between unrelated genus/species (transformation works best when donor and recipient are related). Does this happen in nature? Yes, it can occurs naturally among a few genera of bacteria. In *E. coli* and *Salmonella*, roughly 17% of their genes have been acquired from other speices (over 100 million years). Such ―horizontal transfer‖ is an important issue for the spread of antibiotic resistance and. This process was first demonstrated in *Streptococcus pneumoniae*, by F. Griffith 1928.

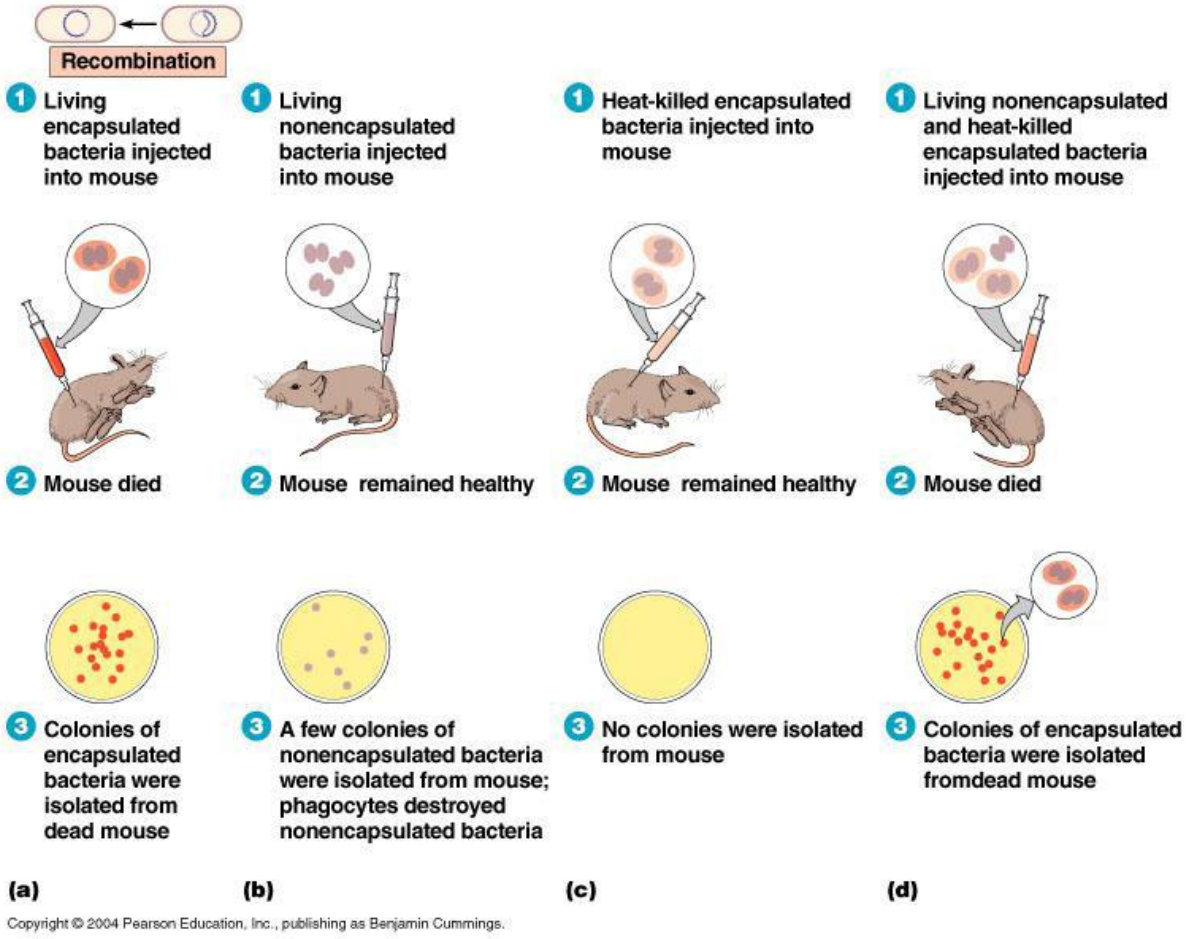


Figure : Griffith‘s experiment: Inject living encapsulated bacteria into mice.

In this experiment, when Griffith Inject living encapsulated bacteria into mice, mice die, and encapsulated bacteria isolated from dead mice. Inject living non-capsulated bacteria into mice, mice remain healthy, a few non-capsulated bacteria can be isolated from the living mice – most phagocytized by leukocytes. Inject heat-killed encapsulated bacteria into mice, mice remain healthy, no bacteria isolated from the living mice. Inject living non-capsulated

and heat-killed encapsulated bacteria into mice, mice die, isolate encapsulated bacteria from dead mice.
Non-capsulated bacteria took up pieces of DNA from dead encapsulated bacteria, some of the pieces contained genes for capsule, a virulent bacteria were transformed by these genes into the encapsulated virulent strain.
The recipient cell must be **competent**, permeable to DNA by alterations in cell wall that allow large molecule like DNA to get through (in lab we use chemical agents to poke holes).These cells can pick up DNA from dead cells and incorporate it into genome by recombination (e.g. antibiotic resistance).

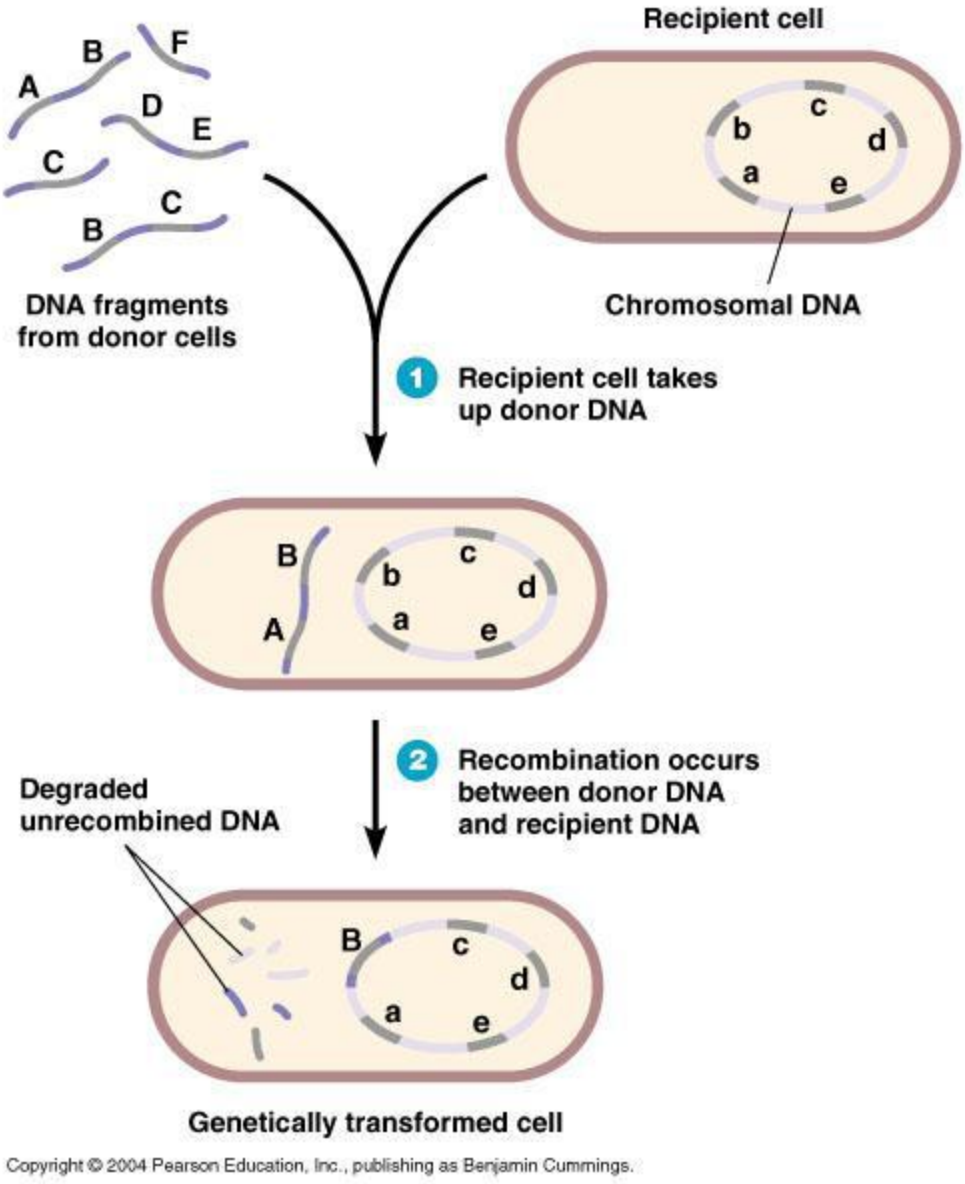


Figure: Transformation process in bacterial cell.

Transformation occurs naturally in many bacteria (e.g. *Bacillus, Streptomyces* and *Haemophillus* spp.) although competence (ability to take up exogenous DNA) is usually transient, being associated with a particular physiological state and requiring the expression of specific competence factors. Other species of bacteria, including *E. coli,* are refractory to natural transformation, but a state of competence can be induced artificially which allows DNA uptake; this has facilitated the use of *E. coli* for molecular cloning .

With the pneumococcus, cells spontaneously become competent to take up DNA. Such naturally-occurring transformation has been most studied in *Bacillus subtilis* and *Haemophilus influenzae* (as well as *S. pneumoniae*) and was for some time thought to be limited to these and related species. It is now known to be much more widespread. In particular, transformation contributes extensively to the antigenic variation observed in the gonococcus (*Neisseria gonorrhoeae*) through the transfer of *pil* genes coding for the major protein subunit of the surface appendages (pili) by which the bacteria attach to epithelial cells. Although the number of species in which natural transformation has been demonstrated is still quite limited, it is likely that it occurs, albeit at a low level, in many other bacteria. The details of the process vary between species, but some generalizations are possible. Competence generally occurs at a specific stage of growth, most commonly in late log phase, just as the cells are entering stationary phase. This may be a response to cell density rather than (or as well as) growth phase. For example, in *Bacillus subtilis,* some of the genes involved in the development of competence are also involved in the early stages of sporulation. The development of competence at this stage is associated not only with nutrient depletion but also with the accumulation of specific secreted products (competence factors) which act via a two component regulatory system to stimulate the expression of other genes required for competence**.**

Following the development of competence, double-stranded DNA fragments bind to receptors on the cell surface, but only one of the strands enters the cell. In some species, the process is selective for DNA from the same species, through a requirement for short species-specific sequences. For example, the uptake of DNA by the meningococcus (*Neisseria meningitidis*) is dependent on the presence of a specific 10-bp uptake sequence. The genome of *N. meningitidis* contains nearly 2000 copies of this sequence, which will only occur infrequently and by chance, in other genomes. Similarly, transformation of *Haemophilus influenzae* is facilitated by the presence of a 29-bp uptake sequence which occurs approximately 1500 times in the genome of *H. influenzae*. These organisms will therefore only be transformed efficiently with DNA from the same species.

Natural transformation is of limited usefulness for artificial genetic modification of bacteria, mainly because it works best with linear DNA fragments rather than the circular plasmid DNA that is used in genetic modification. For introducing foreign genes into a bacterial host, various techniques are used to induce an artificial state of competence. Alternatively, a mixture of cells and DNA may be briefly subjected to a high voltage which enables the DNA to enter the cell (a process known as **electroporation**). Although the mechanisms involved are quite different, they all share the characteristic feature of the uptake of 'naked' DNA by the cells and are therefore also referred to as transformation.

Bacteria can be transformed with DNA extracted from a bacterial virus rather than from another bacterium. This process is known as **transfection.**
If the DNA is from a lytic bacteriophage, transfection leads to virus production and can be measured by the standard phage plaque assay . Transfection is useful for studying the mechanism of transformation and recombination because the small size of phage genomes allows for the isolation of a nearly homogeneous population of DNA molecules. By contrast, in conventional transformation, the transforming DNA is typically a random assortment of chromosomal DNA of various lengths, and this tends to complicate experiments designed to study the mechanism of transformation.

## Transduction in Bacteria

***Transduction*** is transfer of DNA from a donor cell to a recipient cell by a phage (short for **"*bacteriophage*,"** a bacterial virus).There are two type of transduction: ***generalized transduction*** and ***specialized transduction*.** In generalized transduction, any bacterial genes can be transferred. Normally , attachment of ***phag*** to the bacterial cell wall and injection of its DNA into the bacterium will directing the synthesis of new viruses (Figure 25). In **specialized transduction**, only certain bacterial genes are transferred ,The reason for this is that there's only certain places (i.e., certain specific DNA sequences) where the bacteriophage can integrate into the chromosome.

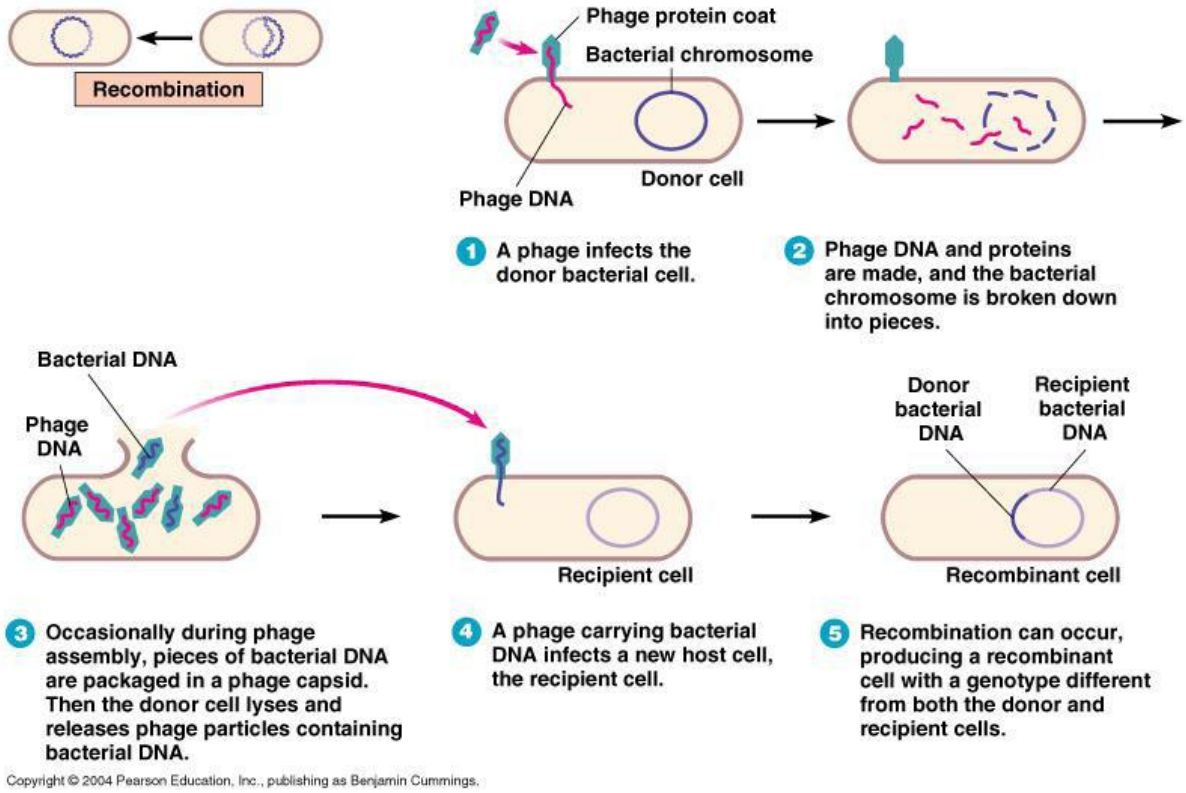


Figure: Bacterial DNA Transfer by Transduction Process.

## Recombination

**Genetic recombination**, the rearrangement of genes from separate groups of genes, usually involves DNA from different organisms; it contributes to genetic diversity. If two chromosomes

break and are rejoined in such a way that some of the genes are reshuffled between the two chromosomes, the process is called ***crossing over*** .In crossing over, genes from two chromosomes are recombined into one chromosome containing some genes from each original chromosome (Figure). However, the ***donor cell*** gives a portion of its total DNA to a different ***recipient cell.*** The recipient, called the ***recombinant,*** has DNA from the donor added to its own Genetic recombination it is DNA exchange of genes between two DNA molecules to form new combinations of genes on chromosome .Genetic recombination contributes to population diversity and it more likely than mutation to provide beneficial change since it tends not to destroy gene function. Generation of recombinant cells is very low frequency event (less than 1%): very few cells in population are capable of exchanging and incorporating DNA. In ***eukaryotes*** recombination during meiosis for sexual reproduction, creates diversity in offspring but parent remains unchanged.

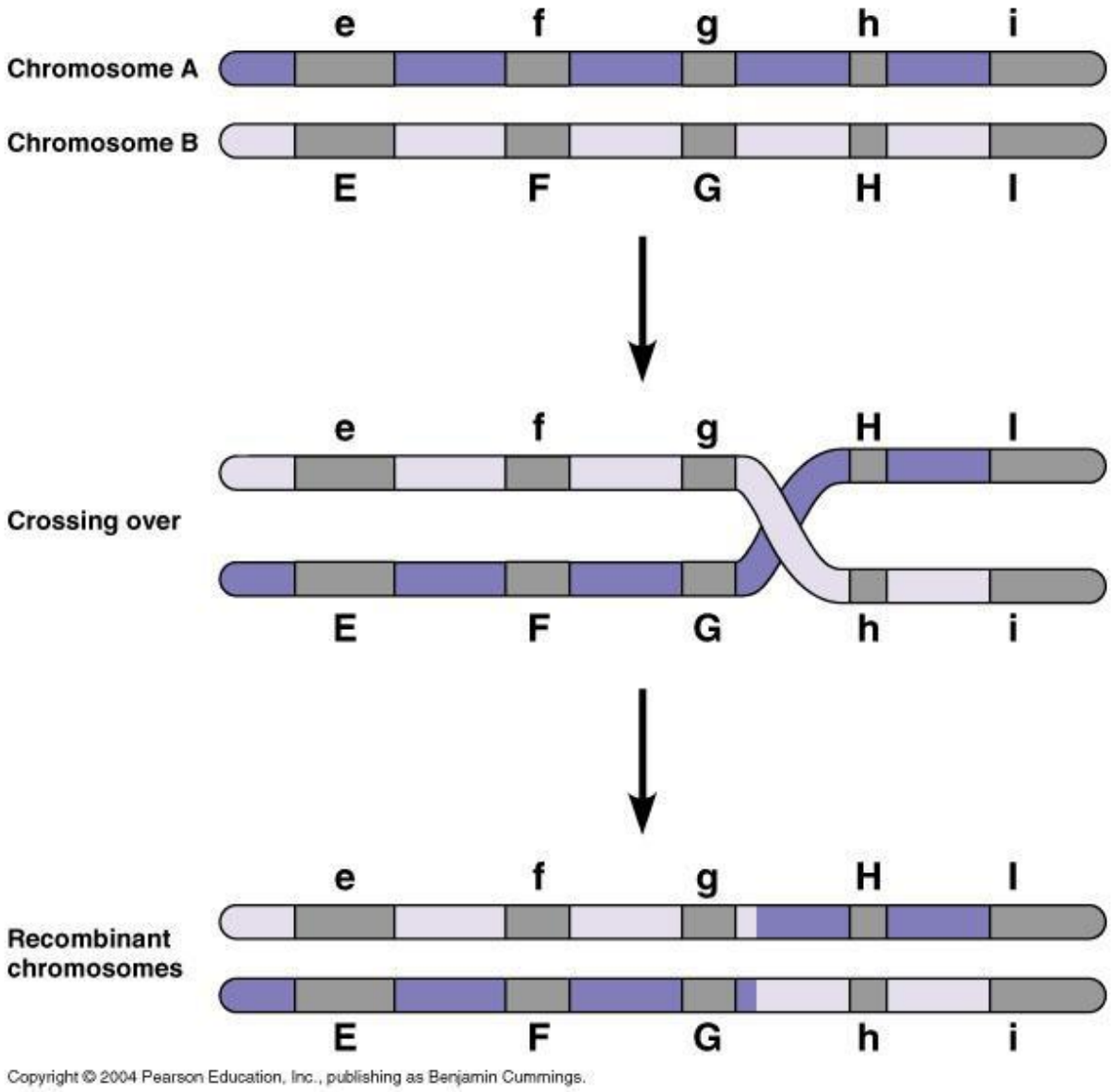



Figure : Genetic recombination and crossing over.