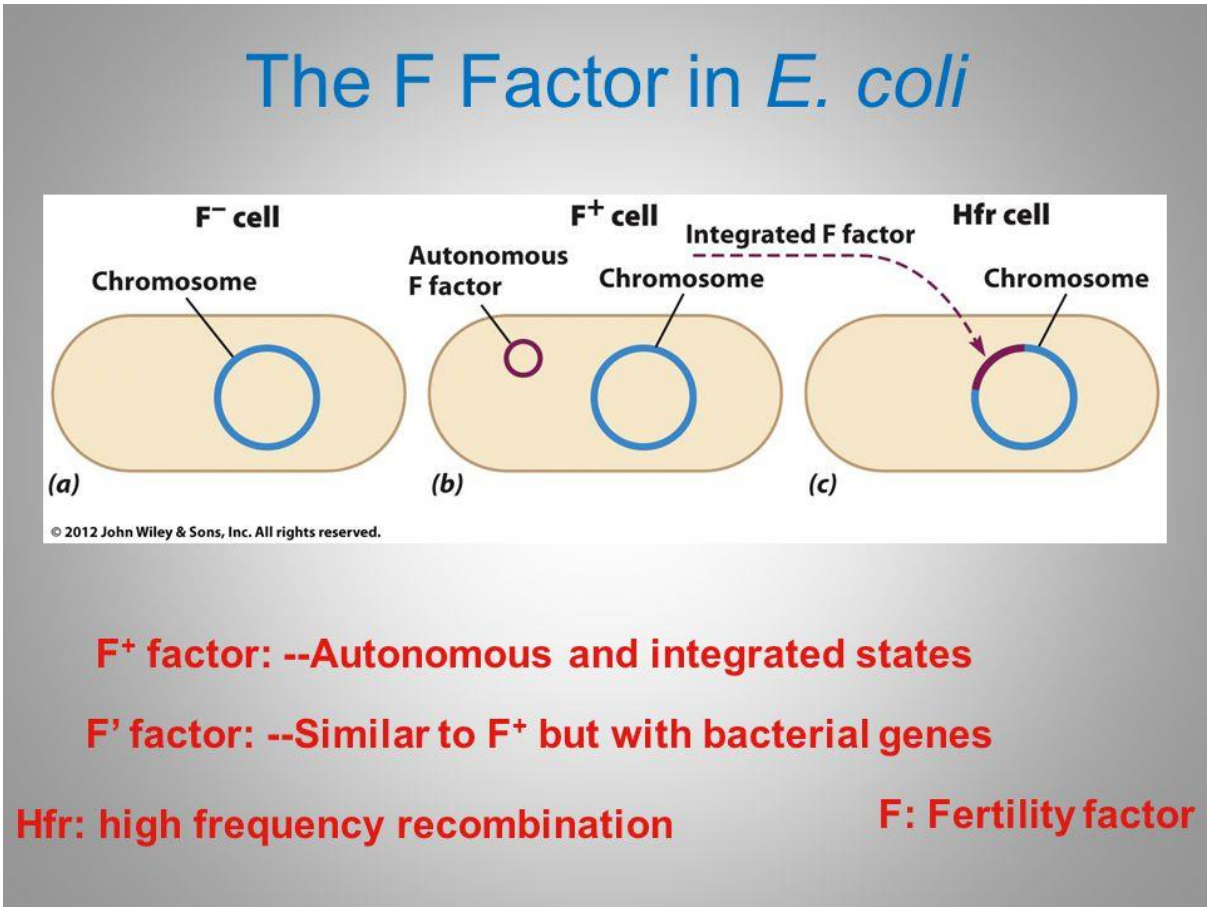
The F plasmid :

is an example of a large plasmid, which contains genes that allow the plasmids DNA to be transferred between cells. It is found in the bacterium E. coli; E. coli containing this F factor are known as F+ and those without are known as F-. The F stands for fertility and the F factor is around 100000 bases in length. The F+ cells have a tube-like structure called a pilus, which allows it to make contact with F- cells. This joining via a pilus in order to transfer DNA between bacteria is known as conjugation. Therefore the F plasmid is known as a conjugative plasmid. Within the E. coli cells, the F plasmid has one or two copies making it a low-copy-number plasmid. During the cell cycle, it replicates once and segregates to both daughter cells.

Transmission of the F plasmid.

Within the F factor are genes, which governs the maintenance and transmission of the F plasmid. As already mentioned, the F plasmid is transferred via conjugation, which occurs due to the pilus known as the F pilus. All the proteins that are associated with the F pilus are transcribed and translated from genes within the F factor. The F plasmid is not transferred to a F- cell via the F pilus, the F pilus merely pulls the two cells together allowing a conjugative junction to form, which contains a pore that allows the DNA to pass from the F+ cell to the F- cell. During the transfer, the F plasmid unwinds and the outer strand breaks, which will be the one that is transferred to the F- cell via the pore in the conjugative junction. Replication of the plasmid then takes place in order to make both single strands of DNA into double-stranded DNA plasmids. In the original F+ cell, the single strand merely undergoes rolling circle replication to once again become double-stranded. In the recipient cell, the linear single-stranded DNA is replicated into a double strand and becomes a circular F plasmid containing the F factor.

Both E. coli cells are now considered to be F+ cells and therefore can both now transfer the F plasmid and therefore the F factor. This transfer only requires a few minutes although is not efficient in natural conditions meaning only 10% of naturally occurring E. coli cells contain the F plasmid and hence the F factor.



Transformation :

is one of the few options for horizontal gene transfer. Though transformation is a natural process, yet only a handful of the organisms are able to perform it naturally. The process of bacterial transformation is also a step of pivotal importance in the field of genetic engineering. The rDNA which is an exogenous DNA, is required to be inserted and expressed in the suitable host. However, majority of the hosts are unable to take up exogenous DNA. Thus, it requires some artificial methods too. The induction of the ability to take up such DNA is called competence. Several methods are being tried since the inception of its concept, but none of them are found to be universal. Therefore, there is a constant requirement of newer methods having advantage and efficiency over the existing ones. The conventional method involves CaCl2treatment followed by heat shock for achieving transformation. There is also employment of device oriented high end methods like electroporation or ultrasound mediated transformation etc. The efficiency of such methods varied widely and is often

specific to a host. Thus, this review is focused on the necessity of transformation and various options that are available to researchers for performing bacterial transformation. It also attempts to strike a comparative study of the existing techniques

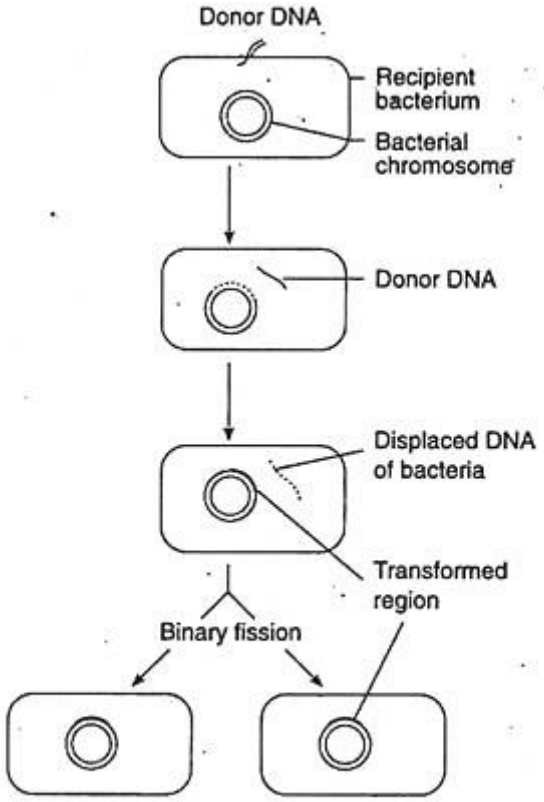


Fig. 2.29 : Diagrammatic representation of Transformation

Transduction:

Transduction is the process by which a virus transfers genetic material from one bacterium to another. Viruses called bacteriophages are able to infect bacterial cells and use them as hosts to make more viruses. After multiplying, these viruses assemble and occasionally remove a portion of the host cell's bacterial DNA. Later, when one of these bacteriophages infects a new host cell, this piece of bacterial DNA may be incorporated into the genome of the new host.

There are two types of transduction: generalized and specialized. In generalized transduction, the bacteriophages can pick up any portion of the host's genome. In contrast, with specialized transduction, the bacteriophages pick up only specific portions of the host's DNA. Scientists have taken advantage of the transduction process to stably introduce genes of interest into various host cells using viruses.