Transformation

It is a kind of genetic recombination where only the carrier of genes, i.e., the DNA molecules of donor cell, pass into the recipient cell through the liquid medium:

It was described by Frederick Griffith (1928), an English bacteriologist. He had done his experiment with laboratory mice and two types of Diplococcus pneumoniae, the pneumonia causing organism. One type has rough (R) non-capsulated cells and another one with smooth (S) capsulated cells. The R-type is non-pathogenic, while the S-type is pathogenic.

The process of transformation is mentioned below (Fig. 2.28):

(i) When live non-pathogenic (R-type) cells are injected in mice, the mice remain alive.

(ii) When dead pathogenic (S-type) cells are injected in mice, the mice also remain alive.'

(iii) When pathogenic (S-type) cells are injected in mice, they suffer from pneumonia and died.

(iv) When live non-pathogenic (R-type) cells are mixed with dead pathogenic (S-type) cells and are injected in mice, they also suffered from pneumonia and died. On isolation of dead tissue of mice, the smooth (S) qapsulated cells are found on agar. The above experiment indicates the conversion of R-type to S-type, called transformation.

Later, James L. Alloway (1932), transformed the rough type cells to smooth type, by using the fragments from dead smooth-type cells and confirmed Griffith's work.

Further, Oswald T. Avery, Colin M. MacLeod and Maclyn N. McCarty (1944) also found that DNA isolated from the fragments could induce the transformation. Their experimental result was the first proof of DNA as the genetic material in living organism. The possible mechanism of transformation can be explained (Fig. 2.29).

The transformation takes place in a few cell of the mixed population. It is an important method of genetic recombination. A few donor cells break apart and an explosive release and fragmentation of DNA take place. A fragment of double stranded DNA (10-20 genes) then gets attached with the recipient cell for entry (Fig. 2.29).

During entry one strand of the fragment becomes dissolved by enzyme leaving the second strand, which then passes to the recipient cell through cell wall and cell membrane.

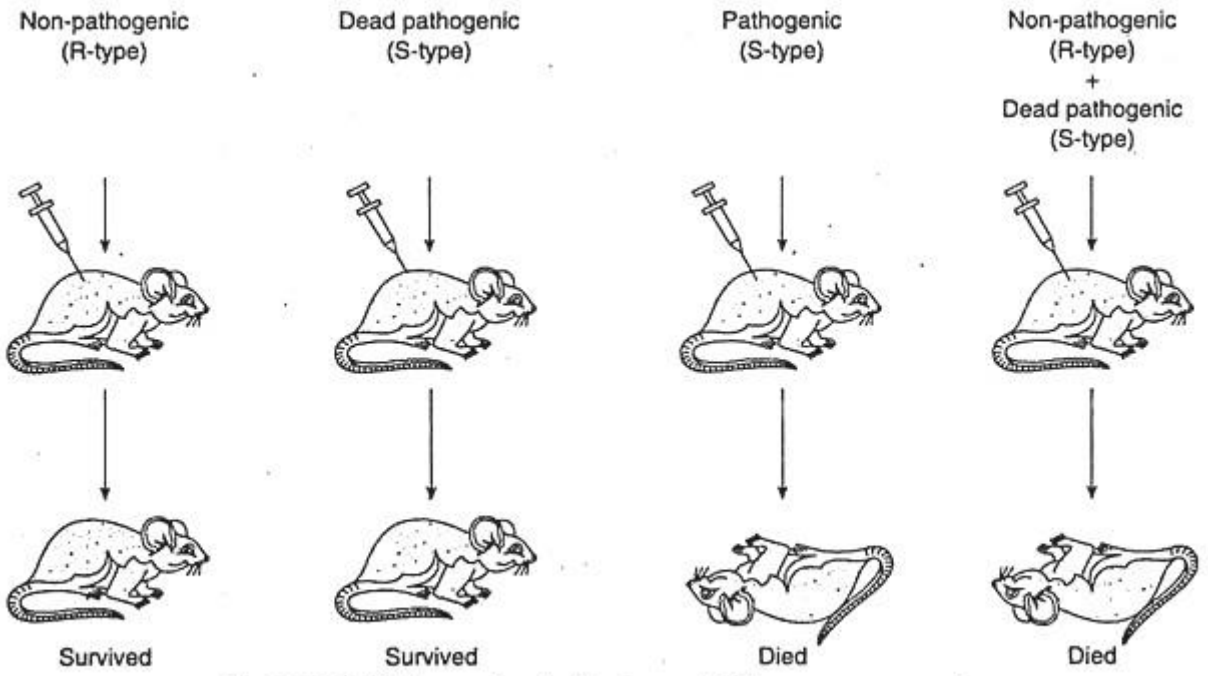

Fig. 2.28 : Griffith's experiment with mice and *Diplococcus pneumoniae*

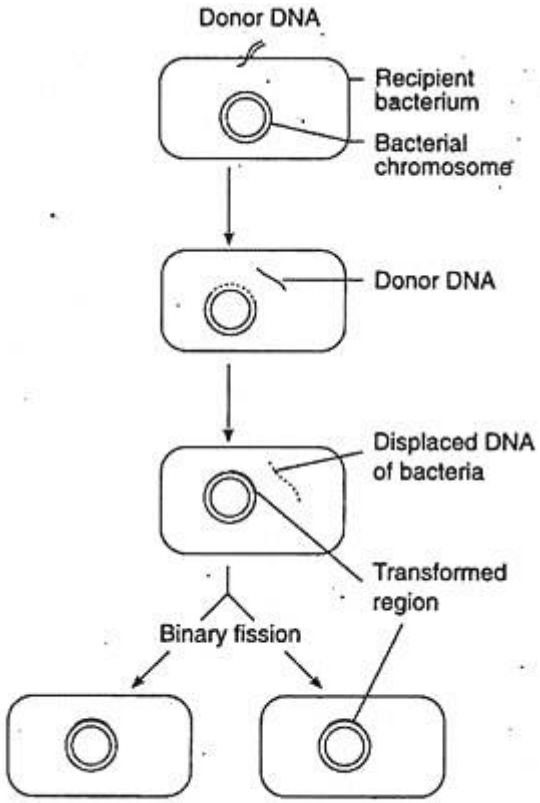

Fig. 2.29 : Diagrammatic representation of Transformation

After entry, a portion of single strand of double stranded DNA of recipient cell gets displaced by enzyme and then replaced by the DNA of donor cell. The displaced DNA is then dissolved by other enzyme. Thus the recipient cell becomes transformed which will display its own as well as the characters of the newly incorporated DNA.

Detailed mechanism of transformation, with special emphasis on natural and induced competence and DNA uptake:

Thus the transformation takes place by horizontal gene transfer through uptake of free DNA by other bacteria. This transformation takes place either spontaneously by taking DNA from the environment, i.e., Natural, or by forced uptake under laboratory condition i.e., Artificial process.

A. Natural Transformation:

During natural transformation, free naked fragments of double stranded DNA of donor cell become attached to the surface of the recipient cell. The free double stranded ON A molecules may be available in the medium by lysis or natural decay of bacteria (Fig. 2.30).

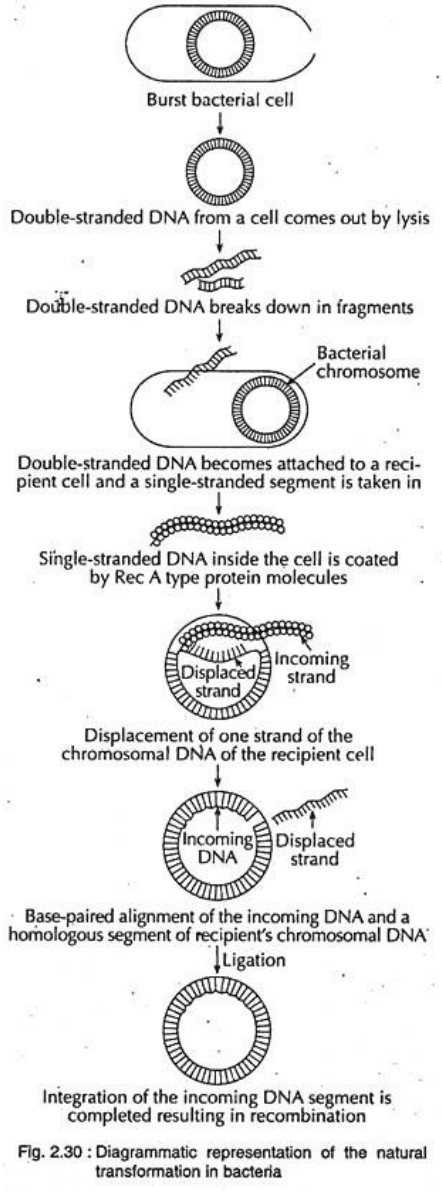

Fig. 2.30 : Diagrammatic representation of the natural transformation in bacteria

After attachment of donor double stranded DNA with the surface of recipient bacterium, one strand is digested by the bacterial nuclease and the remaining one strand is then taken in by an energy-requiring transport system. This uptake of DNA takes place during late logarithmic phase of growth.

During this process, Rec A type of protein plays an important role. The Rec A protein binds with the single stranded DNA and forms a coating around the DNA (Fig. 2.30). The coated single stranded DNA and DNA of recipient cell then move close to each other to get homologous sequence.

After reaching at proper place, the Rec A protein actively displaces one strand of chromosomal DNA of recipient cell. The process requires hydrolysis of ATP to get energy. The incoming DNA strand is then integrated with one strand of bacterial DNA by base pairing and ligation takes place by DNA ligase.

The displaced DNA strand of recipient cell is then digested by cellular DNase activity. Any mismatch between the two strands of new region is corrected by them. Thus the transformation is completed. If the introduced single stranded DNA fails to recombine with the recipient DNA, it is digested by cellular DNase and gets lost.

B. Artificial Transformation:

The E. coli, an ideal material for research is not transformed naturally. Later, it has been discovered that the transformation in E. coli can be done by special physical and chemical treatments. This can be done by exposure of E. coli to high voltage electric field and also by high concentration of $CaCI_2$. Under such condition, the bacterial cells are forced to take up foreign DNA. This type of transformation is called artificial. During this process, the recipient bacterial cells are able to take up double stranded DNA fragments.