

FLUSH END BREAKS

Sometimes RE break Phospho diester bonds between bases that are directly opposite. These are **Even breaks or Flush end breaks**.

PSC 101

Cohen and Boyer in 1973 prepared first Chimaeric DNA using bacterial plasmid. RE used is *E. CORI*. It was also used to cut the DNA of African toad *Xenopus laevis*. **PSC101** is *not found in nature*. It is an *Artificial r DNA* produced in the Lab.

SHOTGUN TECHNIQUE

Used to transfer Animal genes to Plant cells. **Tungsten or Gold coated DNA** is used as Microprojectile. This was done in Wheat and Corn.

ECO REHABILITATION

Use of Genetic Engineering to conserve nature. Control of pollution, production of high yield plants.

T1 PLASMID

Plasmid of *Agrobacterium tumifaciens* introduced in to Tomato, Tobacco, Soyabean

EPSP GENES

Inserted into plants through T1 plasmid to develop Herbicide resistance against *Glyphosate*, a powerful herbicide.

Bt GENE

Gene from *Bacillus thuringiensis* that produce the cry protein which is a natural insecticide against Boll weevil.

ANTIFREEZE GENE

Gene obtained from Antarctic fishes. Transferred to Tomato to reduce ripening.

Nif GENE

Nitrogen fixing gene present in *Rhizobium*

Nod GENE

Nodule forming gene present in *Rhizobium*.

METHODS IN TRANSGENESIS

1. **Electroporation** – Brief electric pulses are applied to create transient micropores in the cell membrane. The technique was discovered by **Wong and Newmann in 1982**.
2. **Lipofection** – Introduction of Artificial gene into a liposome. Useful in transferring large DNA like **Yeast Artificial Chromosome or YAC**.
3. **Micro injection** – Direct transfer of DNA into host nucleus using Micro pipette. Eggs are produced by **Super ovulation** and transferred genes into the eggs by micro injection. The ovum is then transferred into the **Foster mother** already made **Pseudopregnant**.
4. **Retroviral method** – Early cleavage stage embryos (8 cell stage) are infected with RNA virus carrying the transgene. The embryo is then implanted in the uterus of Foster mother.
5. **Embryo stem cell method – Pluripotent – embryonic stem cells** are collected from embryos for genetic engineering. Blastoderm cells are collected and introduced the transgenes by Micro injection. The blastocyst is then introduced in the mother.

TRANSGENIC ANIMALS

First transgenic Mice was produced in 1981 by **Ralph Brinster** and **Richard Palmiter**. Medaka fish, Salmon, Carp etc are used for Transgenesis.

Transgenic Sheep.

Produced to increase Wool production. *Amino acid Serine limit wool synthesis*. In transgenic sheep, the bacterial enzyme converts **Serine to Cysteine**. This increases wool synthesis. Keratin protein of wool also require **Cysteine** for disulphide cross linkages. Transgenic sheep also carries **IX – B Lactoglobulin gene**. *Factor IX is a clotting factor* in the milk.